The American Journal of Medicine

The American Journal of Medicine

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(brand of pentaerythritol tetranitrate)

to relieve the acute attack and sustain coronary vasodilatation

the long-acting emergency tablet for "stress days"

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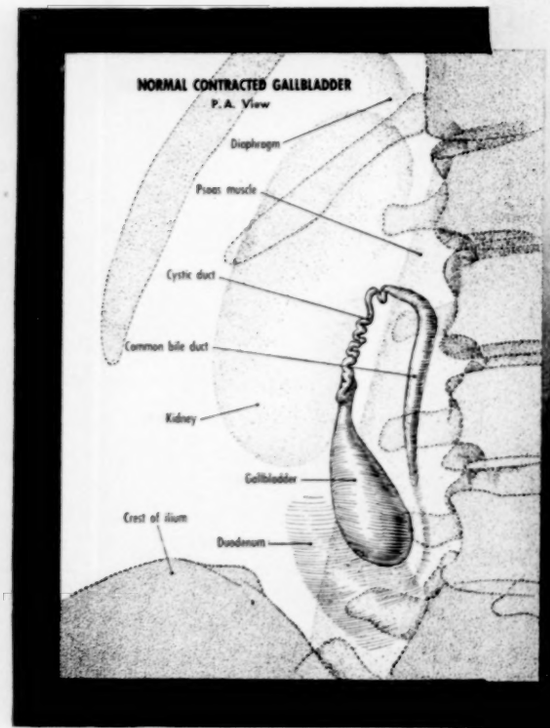

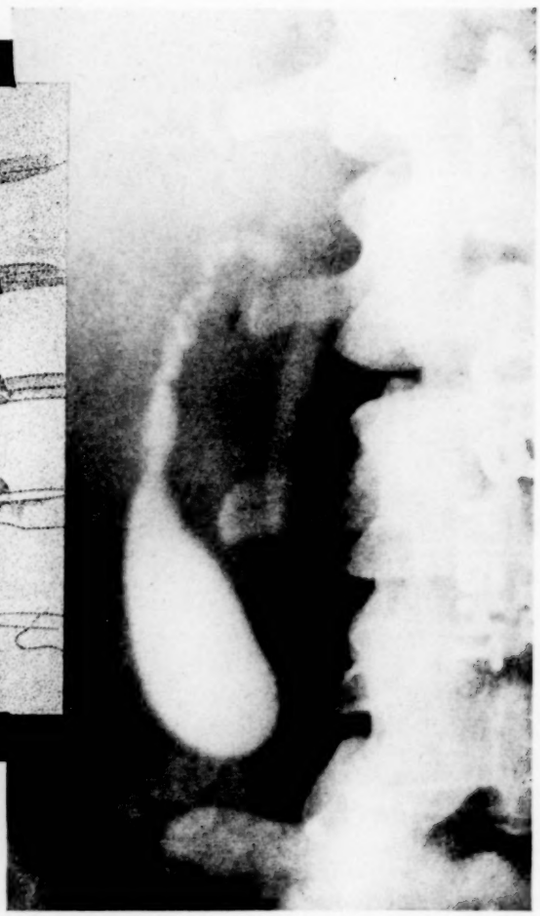

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Buckstein, Jacob: The Digestive Tract in Roentgenology. Philadelphia, J. B. Lippincott Co., 2nd ed., 1953, vol. 2, p. 1003.

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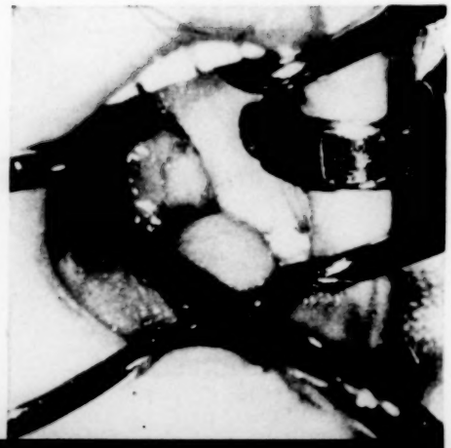
Vol. XXIV APRIL, 1958 No. 4

CONTENTS

Contents continued on page 5

"...providing a cleaner operative field."[1]



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1. Ryan, E. R.: A New Aid to Tonsil and Adenoid Surgery, *Clinical Medicine*, 5, 327 to 331, (March) 1958.

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ORANGE, N. J.

CONTENTS continued–April 1958

VOLUME TWENTY-FOUR NUMBER FOUR

Contents continued on page

CONTENTS continued–April 1958

VOLUME TWENTY-FOUR NUMBER FOUR

Seminar on Liver Disease

Clinico-pathologic Conference

Case Reports

Contents continued on page 9

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to relieve pressures
in your patients

to relieve pressures
on your patients

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Finnerty, F. A. Jr.: New York State J. Med. 57: 2957 (Sept. 15) 1957.

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Corrin, K. M.: Am. Pract. & Dig. Treatment 8: 721 (May) 1957.

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CONTENTS continued–April 1958

VOLUME TWENTY-FOUR NUMBER FOUR

COMING . . . in the May Issue

Symposium on Renal Physiology

GILBERT H. MUDGE, M.D. *and* JOHN V. TAGGERT, M.D.

Guest Editors

Advertising Index on Page 105

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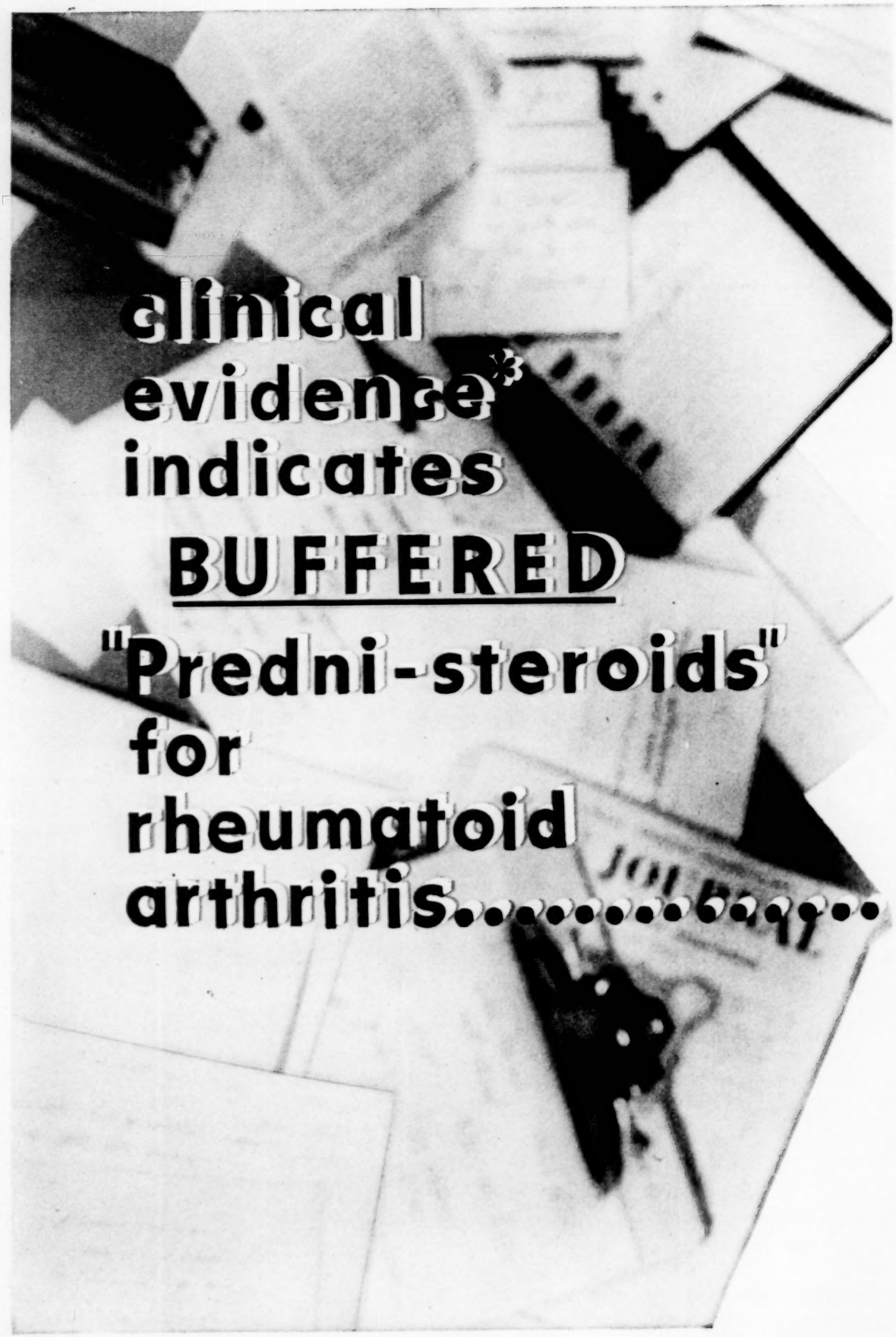
clinical
evidence*
indicates
BUFFERED
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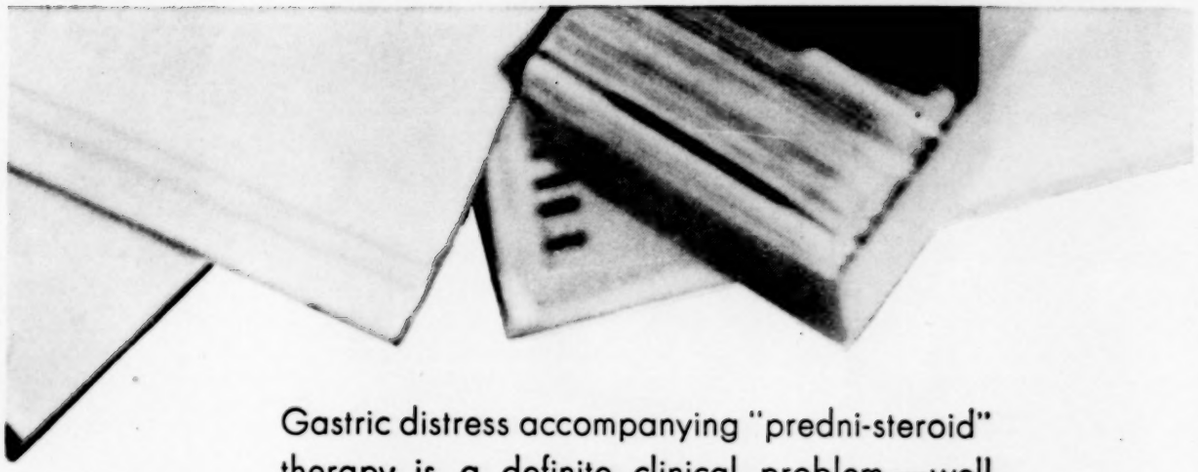
for
rheumatoid
arthritis.....buff...



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*"In view of the beneficial responses observed when antacids and bland diets were used concomitantly with prednisone and prednisolone, we feel that these measures should be employed prophylactically to offset any gastrointestinal side effects."—Dordick, J. R. *et al.*: N. Y. State J. Med. 57:2049 (June 15) 1957.

*"It is our growing conviction that all patients receiving oral steroids should take each dose after food or with adequate buffering with aluminum or magnesium hydroxide preparations."—Sigler, J. W. and Ensign, D. C.: J. Kentucky State M. A. 54:771 (Sept.) 1956.

*"The apparent high incidence of this serious [gastric] side effect in patients receiving prednisone or prednisolone suggests the advisability of routine co-administration of an aluminum hydroxide gel."—Bollet, A. J. and Bunim, J. J.: J. A. M. A. 158:459 (June 11) 1955.

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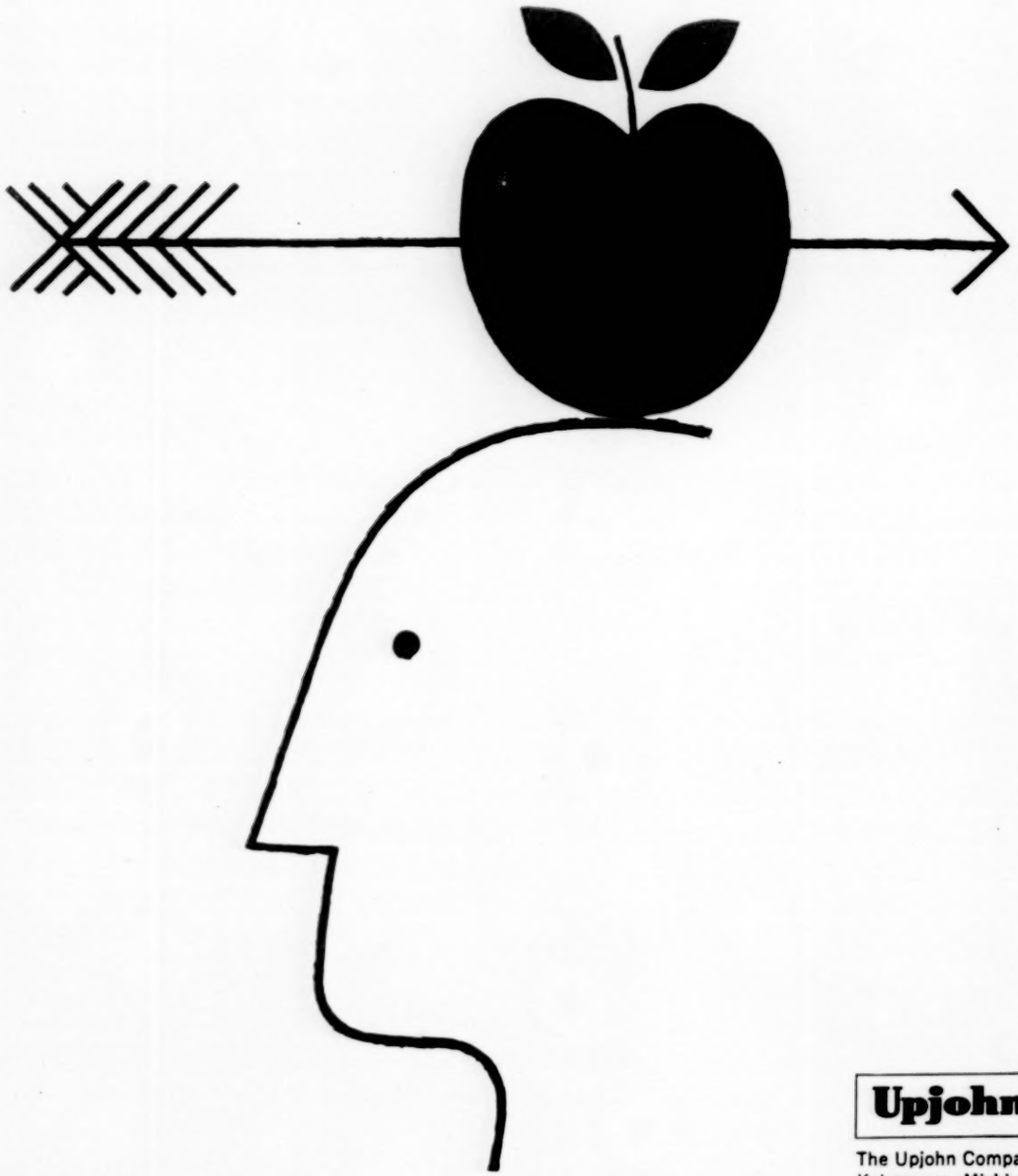

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References: 1. Flodin, N. W.: Am. Miller & Processor *81*:30 (July) 1953. 2. Block, R. J., in Advances in Protein Chemistry, Anson, M. L., and Edsall, J. T., eds., New York, Academic Press, Inc., 1945, vol. 2, p. 119.

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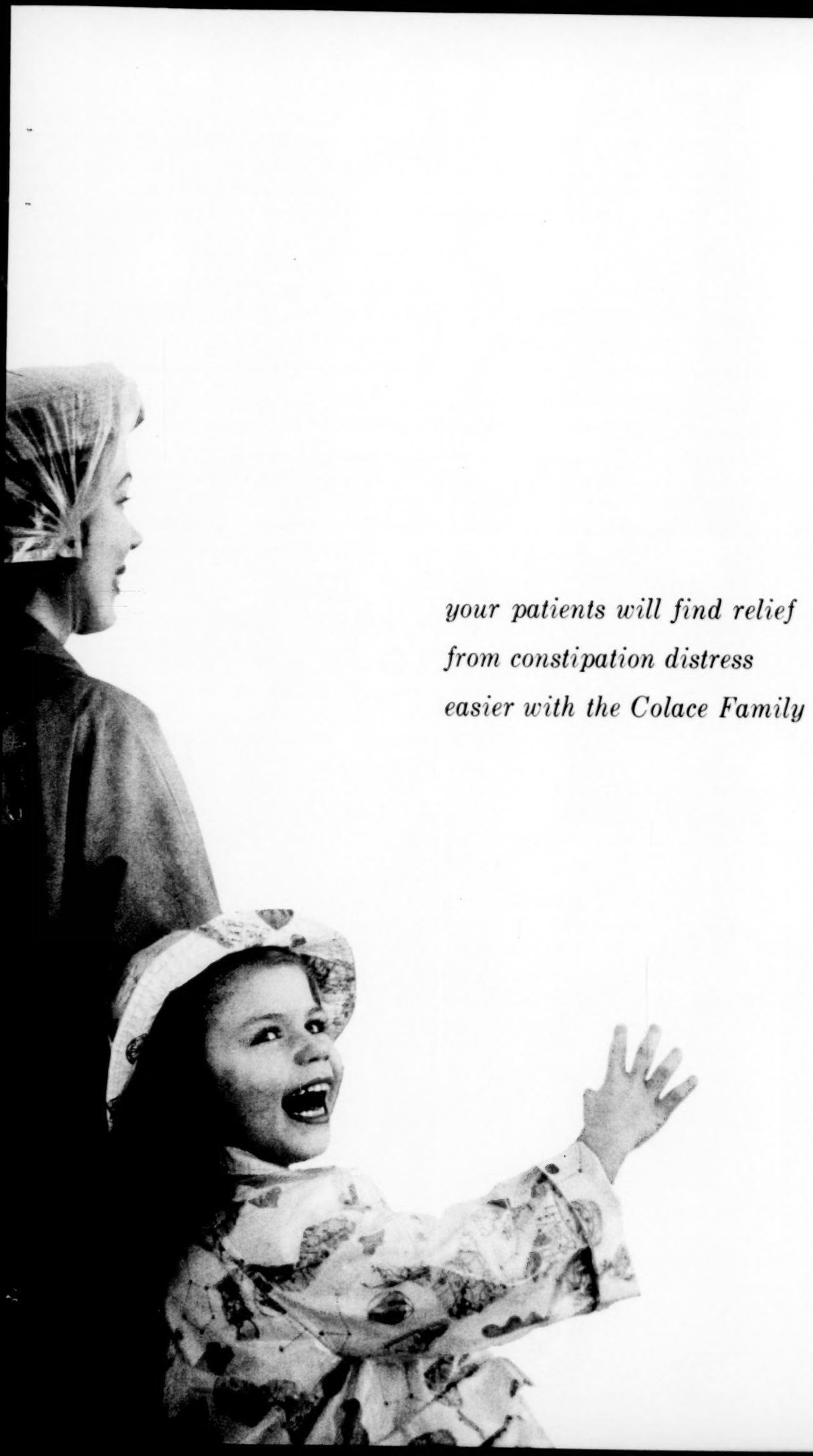

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when defecation is infrequent because bowel motility is inadequate

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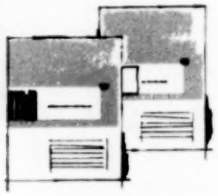

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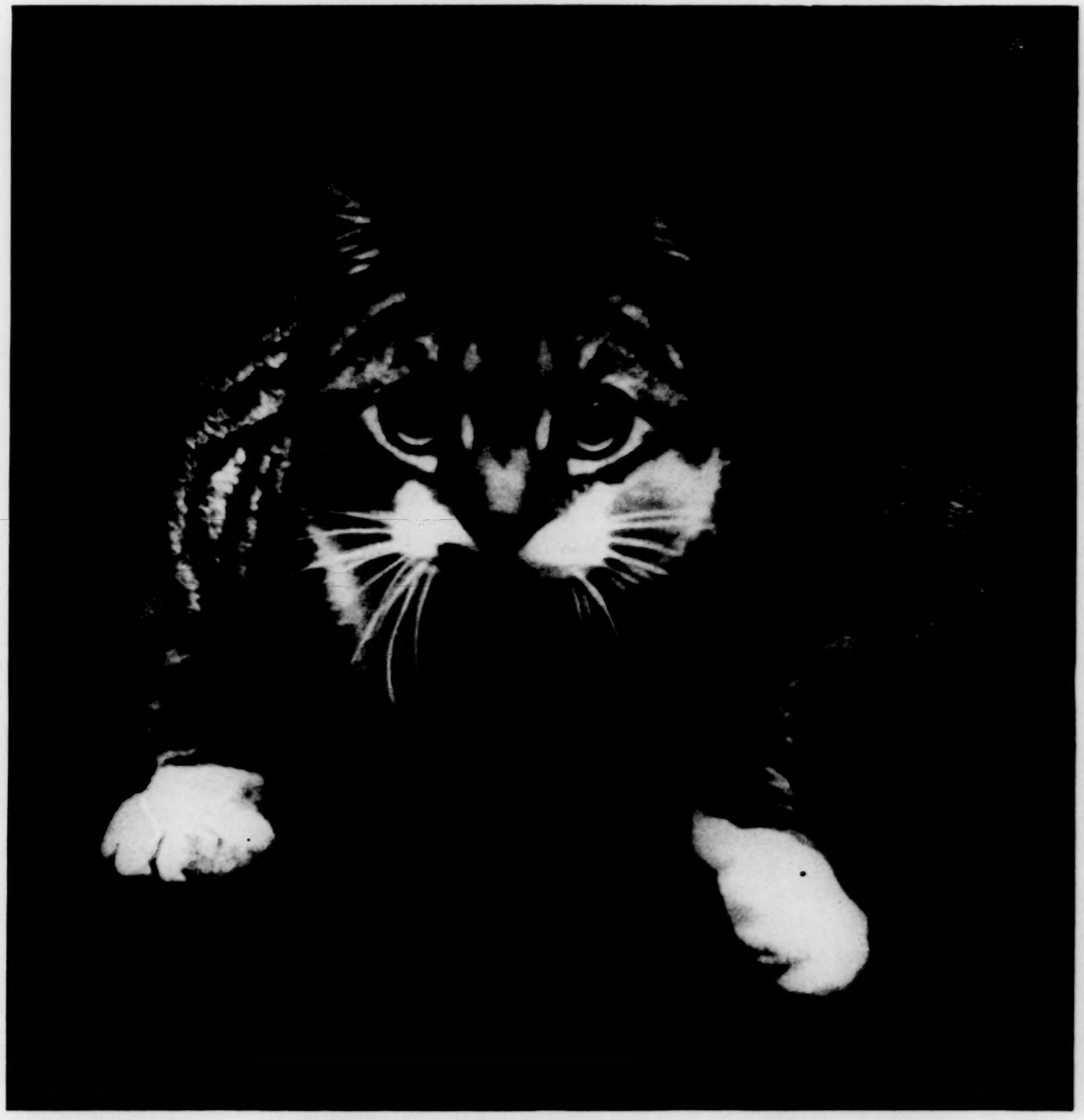

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REFERENCES (1) Woolington, S. S.; Adler, S. J., & Bower, A. G., in Welch, H., & Marti-Ibanez, F.: Antibiotics Annual 1956-1957, New York, Medical Encyclopedia, Inc., 1957, p. 365. (2) Ditmore, D. C., & Lind, H. E.: *Am. J. Gastroenterol.* **28**:378, 1957. (3) Hasenclever, H. F.: *J. Iowa M. Soc.* **47**:136, 1957. (4) Waisbren, B. A., & Strelitzer, C. L.: *Arch. Int. Med.* **99**:744, 1957. (5) Holloway, W. J., & Scott, E. G.: *Delaware M. J.* **29**:159, 1957. (6) Rhoads, P. S.: *Postgrad. Med.* **21**:563, 1957. (7) Petersdorf, R. G.; Bennett, I. L., Jr., & Rose, M. C.: *Bull. Johns Hopkins Hosp.* **100**:1, 1957. (8) Royer, A.: Changes in Resistance to Various Antibiotics of Staphylococci and Other Microbes, paper presented at Fifth Ann. Symp. on Antibiotics, Washington, D. C., Oct. 2-4, 1957. (9) Doniger, D. E., & Parenteau, Sr. C. M.: *J. Maine M. A.* **48**:120, 1957. (10) Josephson, J. E., & Butler, R. W.: *Canad. M. A. J.* **77**:567 (Sept. 15) 1957.

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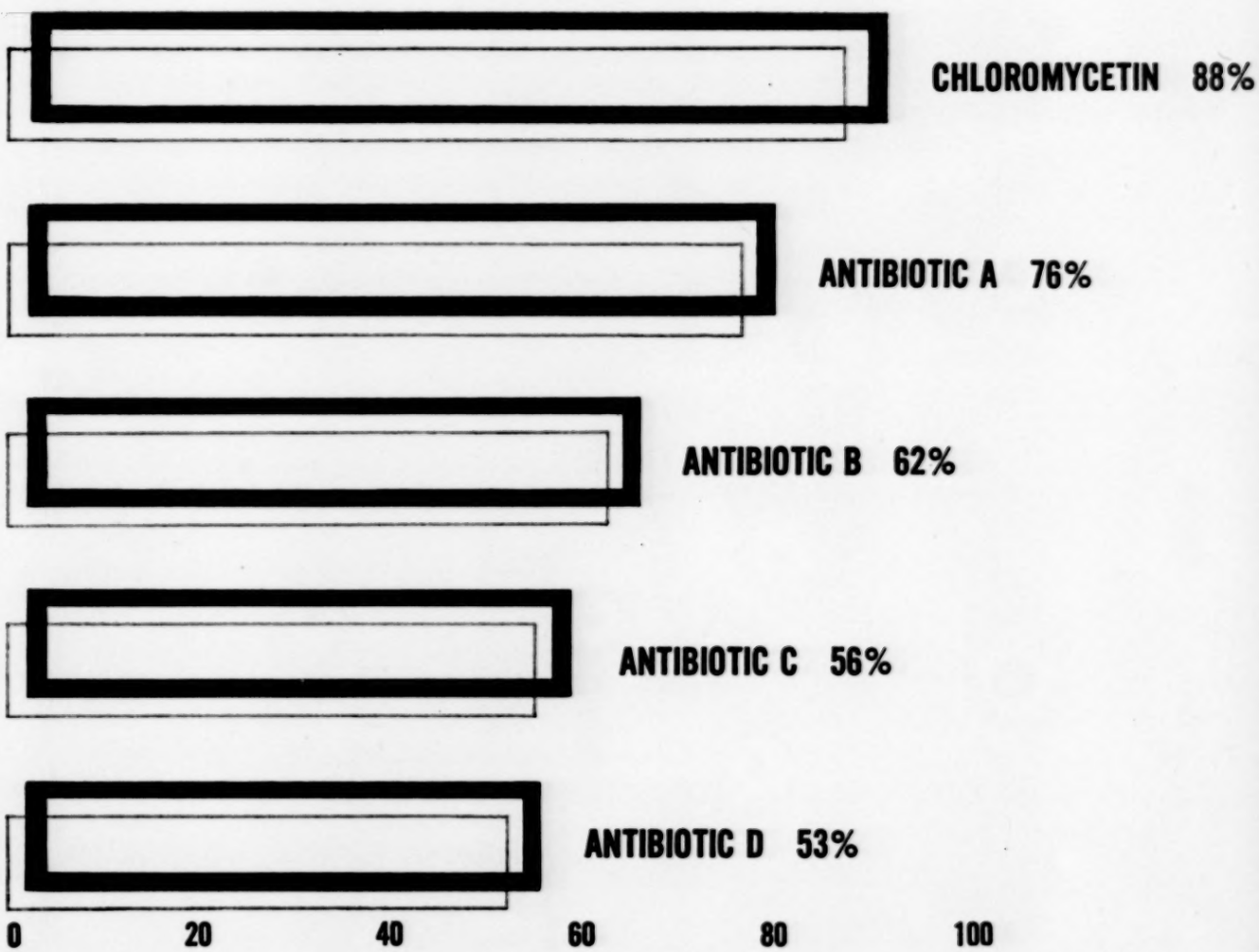

*Adapted from Ditmore and Lind.[3] Organisms tested were isolated from stools of 48 patients.

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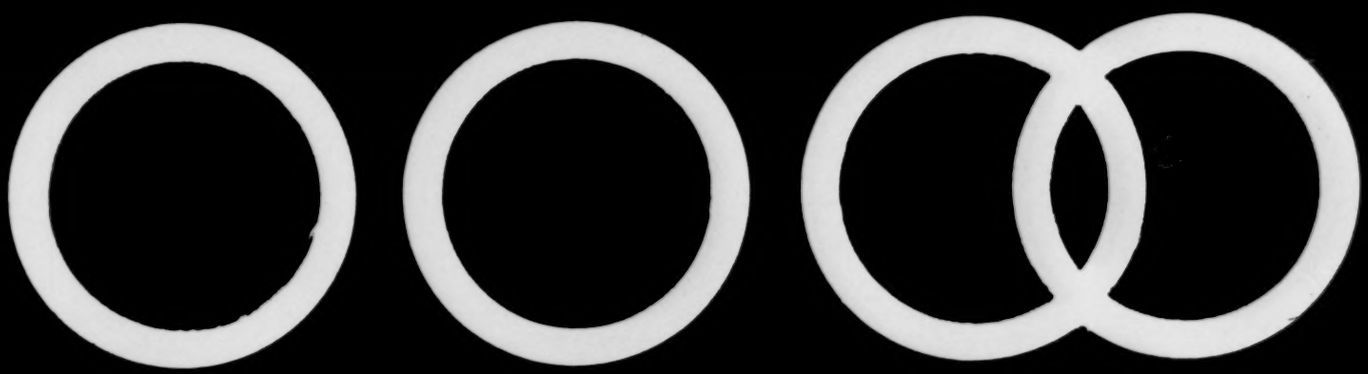

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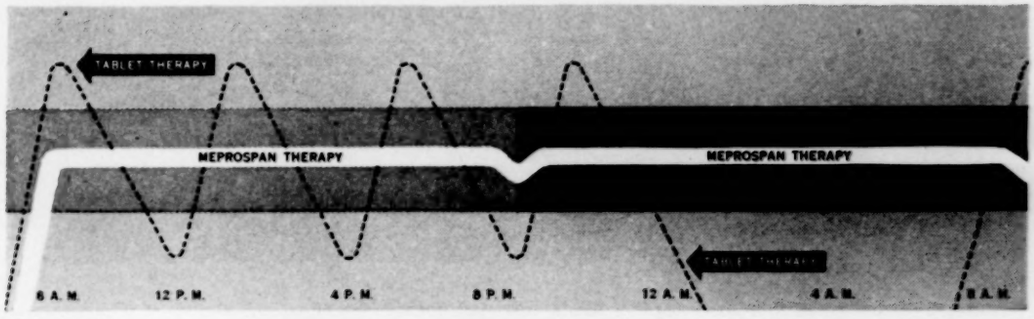

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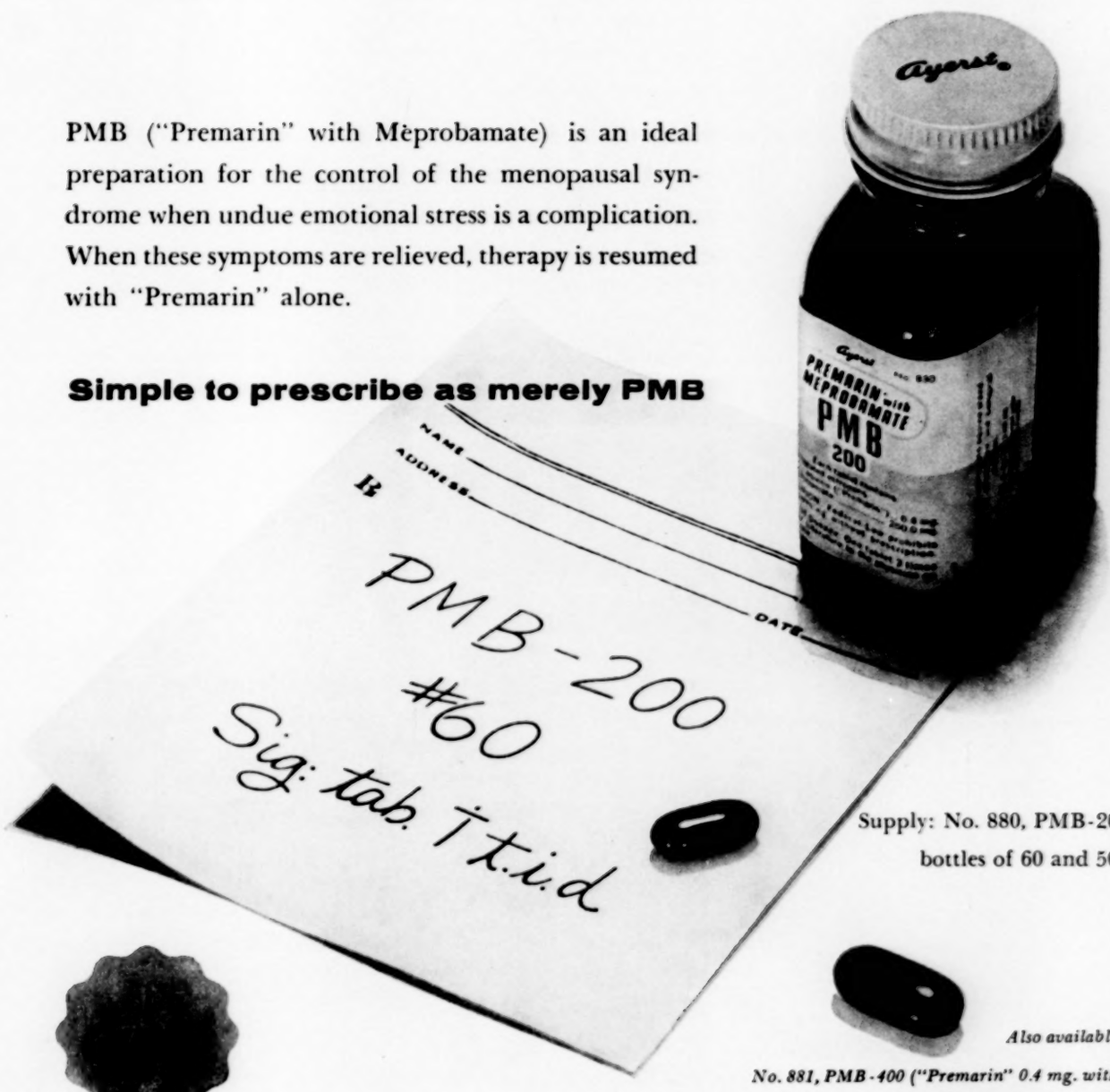

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Rauvera—the combination of alseroxylon and alkavervir—is much more effective than either drug alone. This combination produces "no postural hypotension, no organ toxicity, and no sensitization reactions. Tolerance does not develop on prolonged administration...hypotensive action is steady and prolonged and persists over the entire twenty-four hours."[1] Rauvera therapy can be continued over long periods of time.

"subjective relief...even more so"[1]

Alseroxylon and alkavervir "when combined produce mutual reinforcement so that...more severe cases respond," yet "side effects are minimal."[2] Most patients feel better, are less tired and are free from headaches.[3] Anxiety and tension are relieved...pulse rate slowed...such symptoms as "heart consciousness," tinnitus, vertigo, giddiness and insomnia disappear rapidly —leaving a calm and relaxed patient.

RAUVERA®

1 mg. alseroxylon—3 mg. alkavervir in each scored tablet.

1. Bendig, A.: New York State J. M. 66:2523, 1956. 2. La Barbera, J. F.: Med. Rec. and Ann. 50:242, 1956. 3. Gilchrist, A. R.: Brit. M. J. No. II:1011, 1956.

SMITH-DORSEY • a division of The Wander Company • Lincoln, Nebraska

"Nocturia and orthopnea have disappeared since he's on NEOHYDRIN—and he's edema-free when he wakes in the morning."

oral

organomercurial

diuretic

TABLET
NEOHYDRIN®
BRAND OF CHLORMERODRIN

LAKESIDE

84358

for pain... as effective as codeine without codeine's liabilities

Zactirin*

NON-NARCOTIC

Potently Analgesic

Effectively Anti-inflammatory

2 ZACTIRIN tablets are equivalent in analgesic potency to ½ grain of codeine plus 10 grains of acetylsalicylic acid.

Supplied: Distinctive, 2-layer yellow-and-green tablets, bottles of 48. Each tablet contains 75 mg. of ethoheptazine citrate and 325 mg. (5 grains) of acetylsalicylic acid.

Wyeth®
Philadelphia 1, Pa.

to minimize
morning joint stiffness...

PERSISTIN*

Night-long salicylate therapy with a single dose of Persistin at bed-time helps prevent "joint jelling" in arthritic patients.

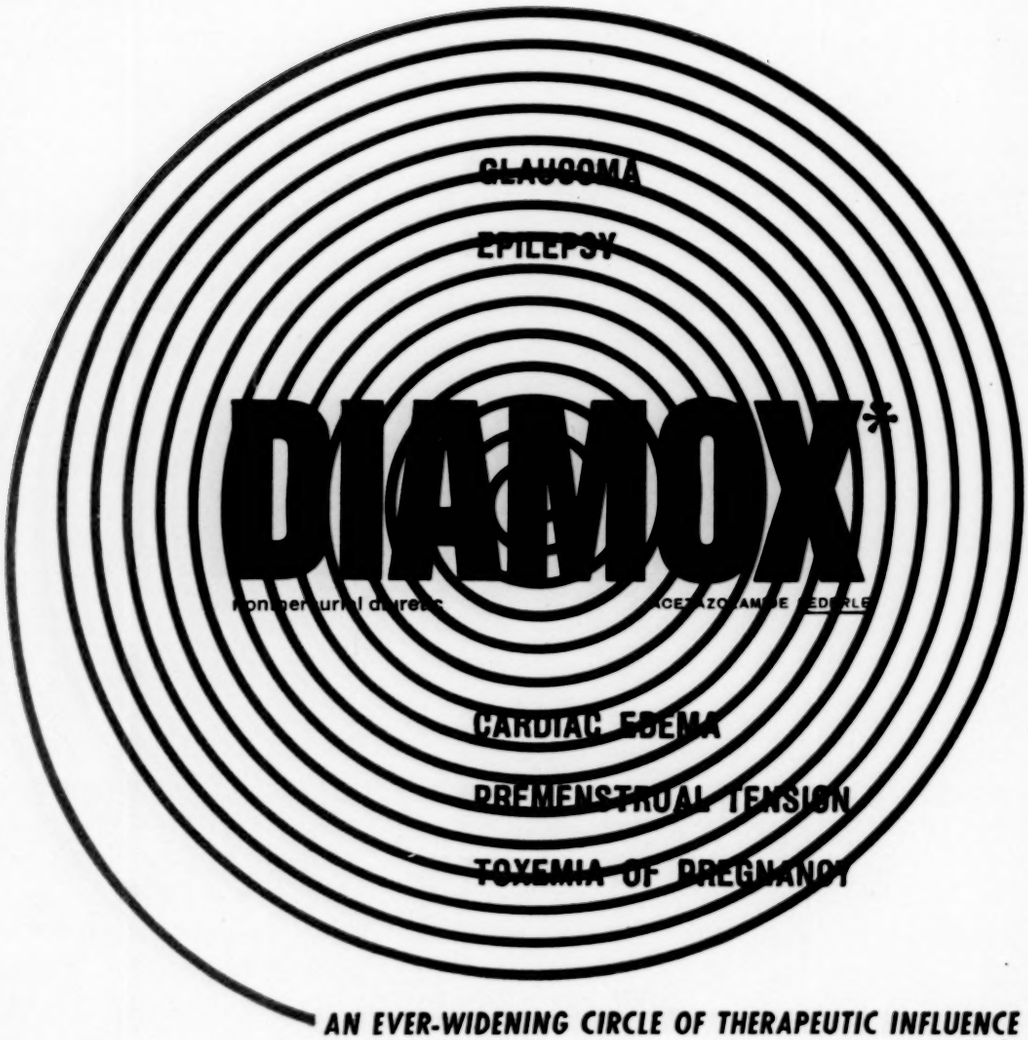
Each Persistin tablet contains acetylsalicylic acid 2½ gr. (160 mg.) and salicylsalicylic acid 7½ gr. (480 mg.). The latter ingredient is slowly absorbed and eliminated for prolonged salicylate action up to 8 hours.

Complete dosage information in PDR . . . bottles of 90 tablets

Samples and literature on request

Sherman Laboratories
Detroit 11, Michigan

*Trademark–Pat. Pend.



AN EVER-WIDENING CIRCLE OF THERAPEUTIC INFLUENCE

A versatile, dependable **diuretic** – the most widely prescribed of its kind: its unique action as a carbonic anhydrase inhibitor has proved strikingly effective not only in conditions requiring diuretic treatment but in the management of other conditions as well.

DIAMOX is well-suited to long-term therapy. Low toxicity, freedom from renal and gastro-intestinal irritation, ease of administration make its use simple and singularly free of complications. Excretion of the drug by the kidney is complete within 24 hours, with no cumulative effects.[1]

Diuretic treatment with DIAMOX results in continuous rather than intermittent control of edema since DIAMOX is effective in the mobilization of edema fluid and in the prevention of fluid accumulation.[1] A single oral dose, active for 6-12 hours, provides the basis for the highly desirable advantage of daytime diuresis and nighttime rest.

Supplied: Scored Tablets of 250 mg.; Syrup containing 250 mg. per 5 cc. teaspoonful; Vials of 500 mg. for parenteral use.

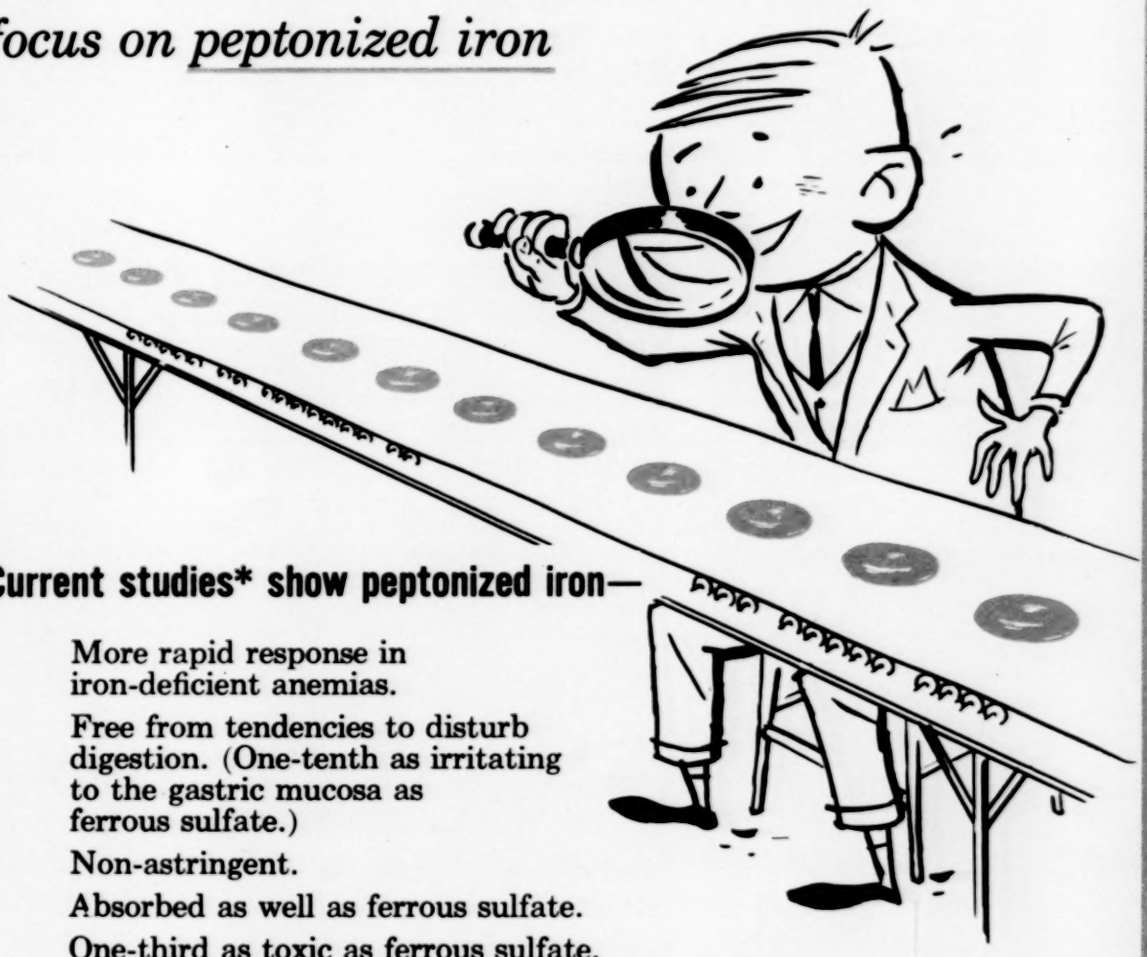
1. Goodman, L. S. and Gilman, A.: The Pharmacological Basis of Therapeutics. Ed. 2. The Macmillan Co., New York, 1955, p. 856.

LEDERLE LABORATORIES DIVISION, AMERICAN CYANAMID COMPANY PEARL RIVER, NEW YORK Lederle

*Reg. U. S. Pat. Off.

For predictable therapeutic advantages . . .

focus on peptonized iron



Current studies* show peptonized iron—

More rapid response in iron-deficient anemias.

Free from tendencies to disturb digestion. (One-tenth as irritating to the gastric mucosa as ferrous sulfate.)

Non-astringent.

Absorbed as well as ferrous sulfate.

One-third as toxic as ferrous sulfate.

LIVITAMIN®

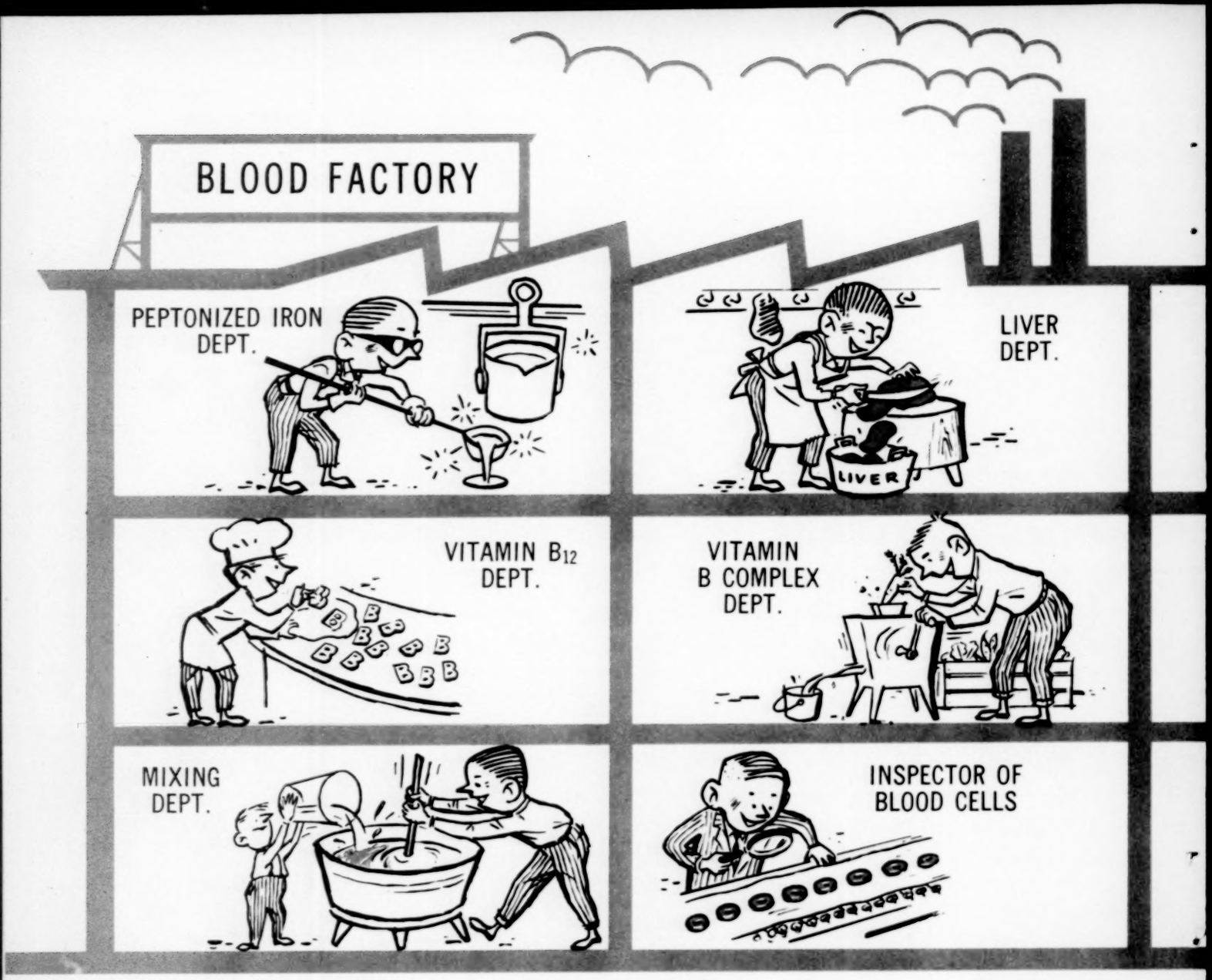
Currently, mailings will be forwarded only at your request. Write for samples and literature.

*Keith, J.H.: Utilization and Toxicity of Peptonized Iron and Ferrous Sulfate, Am. J. Clin. Nutrition *1*:35 (Jan.-Feb., 1957).

The S. E. MASSENGILL Company

BRISTOL, TENNESSEE

NEW YORK • KANSAS CITY • SAN FRANCISCO



The preferred hematinic with PEPTONIZED *iron*

LIVITAMIN®

Peptonized iron is virtually predigested. It is absorbed as well as ferrous sulfate, and is one-tenth as irritating to the gastric mucosa. Anemias refractory to other forms of iron will often respond promptly to Livitamin therapy.

The Livitamin formula, containing the B complex, provides integrated therapy to correct the blood picture, and to improve appetite and digestion.

Each fluidounce contains:

Iron peptonized (Equiv. in elemental iron to 71 mg.)	420 mg.
Manganese citrate, soluble	158 mg.
Thiamine hydrochloride	10 mg.
Riboflavin	10 mg.
Vitamin B_{12} Activity (derived from Cobalamin conc.)	20 mcg.
Nicotinamide	50 mg.
Pyridoxine hydrochloride	1 mg.
Pantothenic acid	5 mg.
Liver fraction 1	2 Gm.
Rice bran extract	1 Gm.
Inositol	30 mg.
Choline	60 mg.

The S. E. MASSENGILL Company

BRISTOL, TENNESSEE

NEW YORK • KANSAS CITY • SAN FRANCISCO

Documentary Case History . . .

Hypertension controlled for four years with Serpasil®

(reserpine CIBA)

K. C., a 67-year-old retired shirt manufacturer, had a 16-year history of hypertension, was troubled by recurrent dizzy spells and headaches. "I'd get several attacks a day. . . . Usually I'd go into the bedroom and lie down." Serpasil therapy was started four years ago, effecting a gradual reduction of the patient's initial blood pressure of 220/120 mm. to the present 140/80. Now well and asymptomatic, ". . . I'm able to go to matinees and see some of the TV shows."

SUPPLIED: TABLETS, 4 mg. (scored), 2 mg. (scored), 1 mg. (scored), 0.25 mg. (scored) and 0.1 mg.

ELIXIRS, 1 mg. and 0.2 mg. Serpasil per 4-ml. teaspoon.

PARENTERAL SOLUTION: *Ampuls*, 2 ml., 2.5 mg. Serpasil per ml. *Multiple-dose Vials*, 10 ml., 2.5 mg. Serpasil per ml.

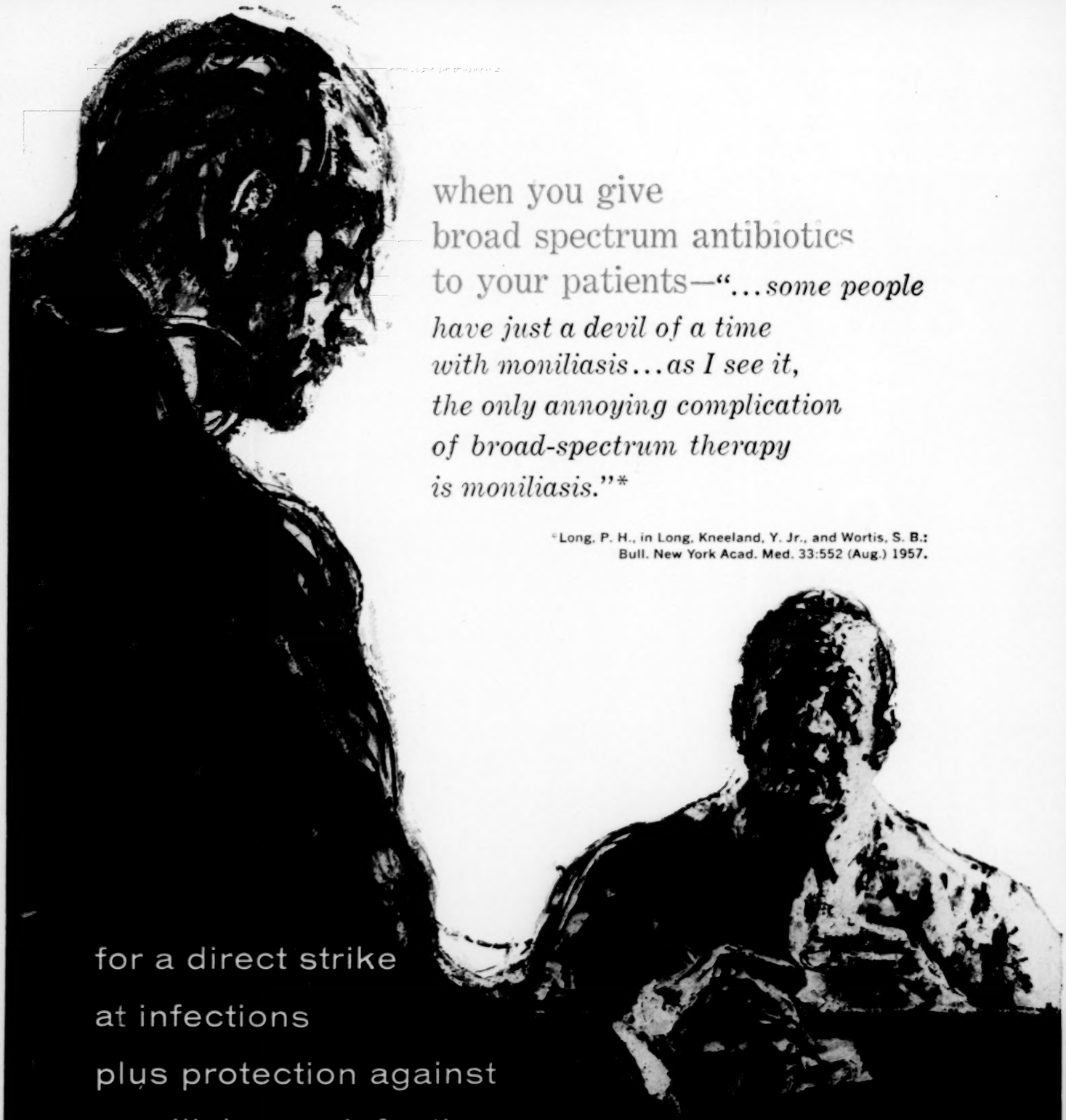
Hypertension controlled through

SYMPATHETIC REGULATION

Serpasil shields the psychic and somatic reaction centers from emotional and environmental stress stimuli, thereby inhibiting the discharge of vasoconstrictive impulses through the sympathetic nerves.

Adapted from Moyer, J. H., Dennis, E., and Ford, R.: Arch. Int. Med. 96:530 (Oct.) 1955.

CIBA SUMMIT, N. J. 2/2535HX



when you give
broad spectrum antibiotics
to your patients—"*...some people*
have just a devil of a time
with moniliasis...as I see it,
the only annoying complication
of broad-spectrum therapy
is moniliasis."*

*Long, P. H., in Long, Kneeland, Y. Jr., and Wortis, S. B.:
Bull. New York Acad. Med. 33:552 (Aug.) 1957.

for a direct strike
at infections
plus protection against
monilial superinfection
the best broad spectrum
antibiotic to use is

THESE ARE YOUR PATIENTS WHO MAY HAVE "JUST A DEVIL OF A TIME WITH MONILIASIS"

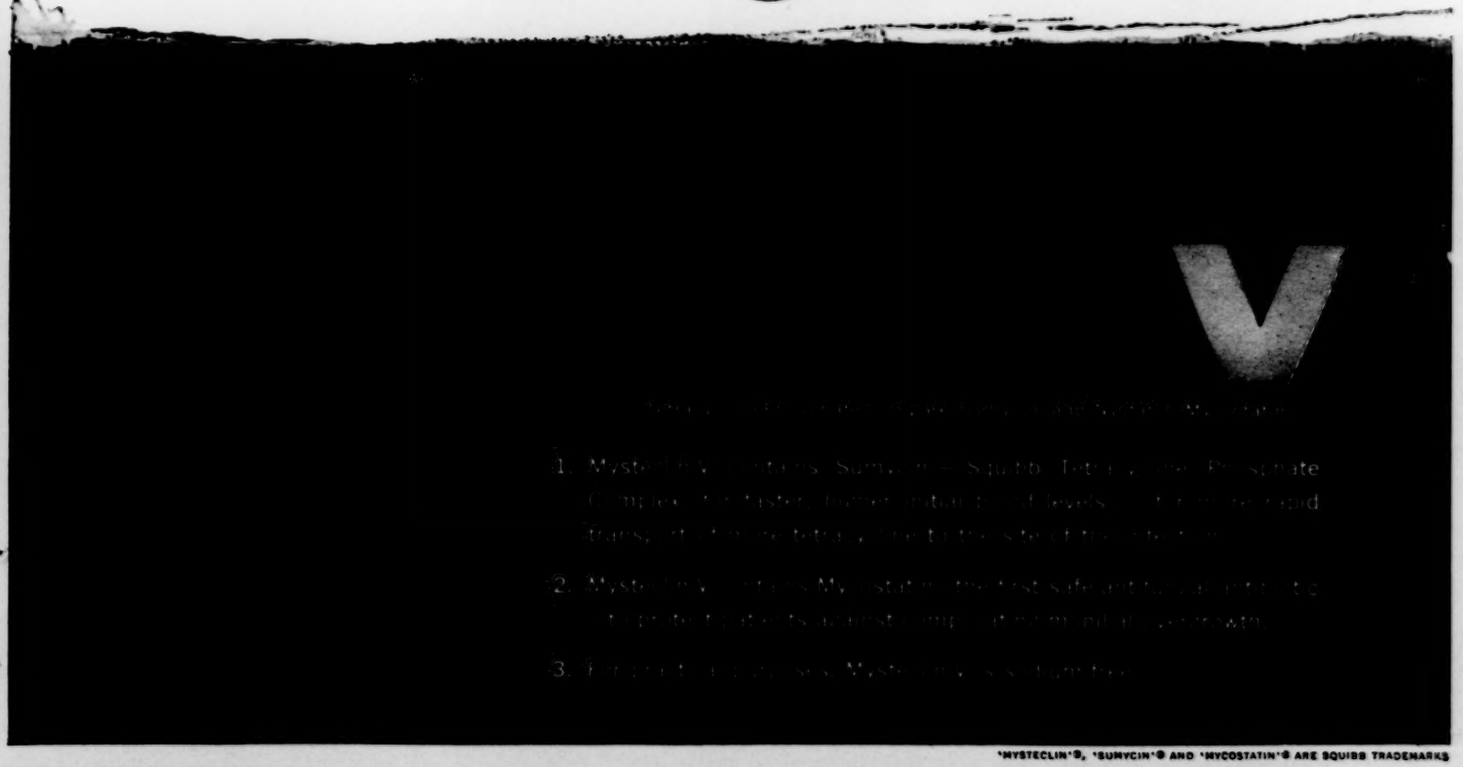
- debilitated patients
- elderly patients
- diabetics
- infants, especially prematures
- those who developed moniliasis on previous broad spectrum therapy
- patients on prolonged and/or high dosage antibiotic therapy
- women, especially when pregnant or diabetic

Mysteclin-V provides you with a dosage form for every clinical need:

	Tetracycline phosphate complex equiv. tetracycline HCl (mg.)	Mycostatin (units)	Packaging
Capsules (per capsule)	250	250,000	Bottles of 16 and 100
Half-Strength Capsules (per capsule)	125	125,000	Bottles of 16 and 100
Suspension (per 5 cc.)	125	125,000	60 cc. bottles
Drops (per cc.—20 drops)	100	100,000	10 cc. dropper bottles

SQUIBB 

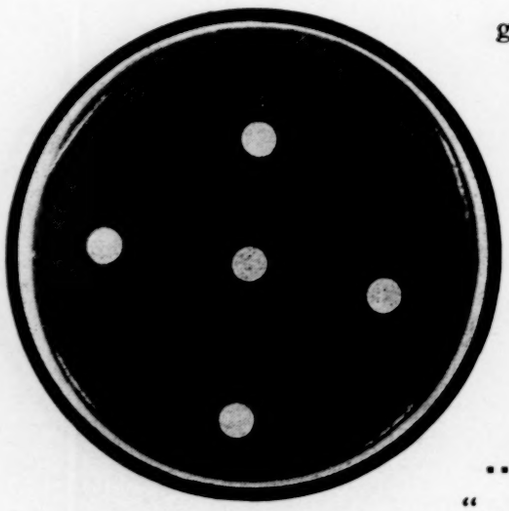
SQUIBB QUALITY—THE PRICELESS INGREDIENT



on the problem of

A POINT OF VIEW IN '55

"At this time, it appears that the problem of antibiotic-resistant bacteria is the greatest fear in the future with chronic infections of the . . . urinary tract . . ."[1]



A POINT OF FACT IN '58

". . . This prediction has proved to be correct for both gram-positive and gram-negative organisms."[2]

. . . WITH ONE NOTABLE EXCEPTION

". . . studies indicate that microorganisms, in vitro and in vivo, do not appear to develop resistance to FURADANTIN."[3]

antibiotic-resistant bacteria

for acute and chronic genitourinary tract infections

FURADANTIN®

brand of nitrofurantoin

AVERAGE FURADANTIN DOSAGE: In acute, complicated or refractory cases and in chronic infections—100 mg. q.i.d., with meals and with food or milk on retiring.

REFERENCES: 1. Flippin, H. F.: Virginia M. Month. 82:435, 1955. 2. Caswell, H. T., et al.: Surg. Gyn. Obst. 106:1, 1958. 3. Nesbitt, R. E. L., Jr., and Young, J. E.: Obst. Gyn., N. Y. 10:89. 1957.

NOW, for hospitalized patients, for severe urinary tract infections when peroral administration of FURADANTIN is not feasible and for serious infections as septicemia (bacteremia): **FURADANTIN** *Intravenous Solution*

NITROFURANS . . . a *new* class of antimicrobials . . . neither antibiotics nor sulfonamides

EATON LABORATORIES, NORWICH, NEW YORK

pattern of subclinical hypothyroidism

pattern for PROLOID®

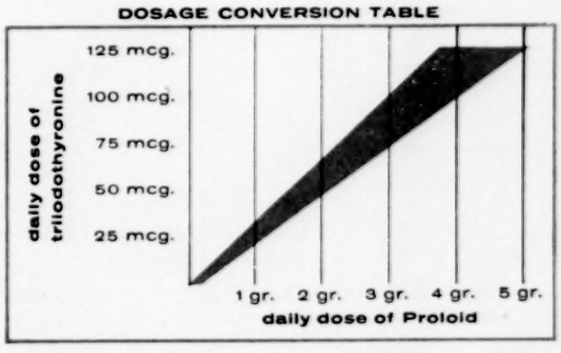
Mild hypothyroidism, a *typical* syndrome is often manifested by *atypical* symptoms. Singly, the complaints of all these patients could be assigned to a variety of conditions. But as you trace the one cause common throughout the group—the typical pattern of mild hypothyroidism emerges.

Often one symptom may supply the clue. A patient's paresthesia, a woman's brittle nails, a child's slow teething may first arouse suspicion. With further investigation, other symptoms fall in line and suspicion may lead to diagnosis.

For final confirmation the physician may turn to the serum PBI test; or, occasionally to empiric confirmation by "break-through" therapy with triiodothyronine. But even when the existence of a hypometabolic state has thus been established (by administration of a synthetic thyroid fraction) long-range thyroid substitution is maintained most successfully and safely with a preparation supplying *all* thyroid fractions in their naturally occurring proportion.

Proloid, the total thyroid complex, fits this specification perfectly. Proloid is standardized both chemically and biologically for metabolic potency. It assures a smooth predictable clinical response.

Five Proloid tablet sizes give precision and convenience on every dosage level—for the maintenance of euthyroidism throughout the patient's lifetime. Proloid is prescribed in the same dosage as ordinary thyroid and is available in ¼, ½, 1, 1½ and 5 grain tablets as well as powder.



PROLOID® *the total thyroid complex*

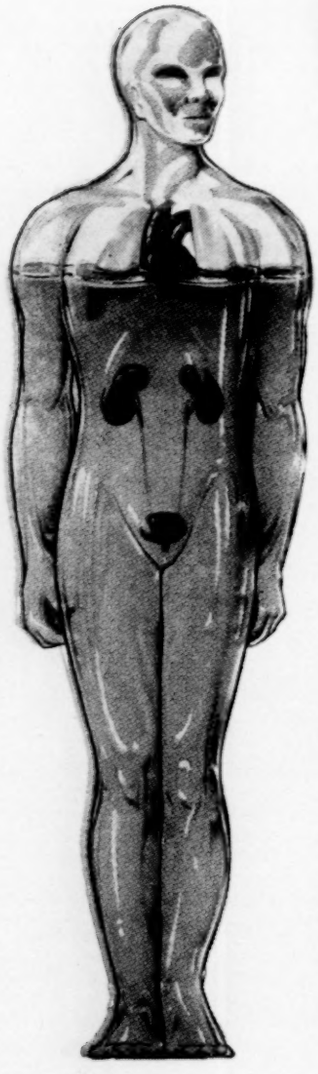
WARNER-CHILCOTT

'DIURIL'

(CHLOROTHIAZIDE)

in

EDEMA



Start therapy with one or two 500 mg. tablets of 'DIURIL' once or twice a day.

BENEFITS:

- The only orally effective nonmercurial agent with diuretic activity equivalent to that of the parenteral mercurials.
- Excellent for initiating diuresis and maintaining the edema-free state for prolonged periods.
- Promotes balanced excretion of sodium and chloride—without acidosis.

Any indication for diuresis is an indication for 'DIURIL':

Congestive heart failure of all degrees of severity; premenstrual syndrome (edema); edema and toxemia of pregnancy; renal edema—nephrosis; nephritis; cirrhosis with ascites; drug-induced edema. May be of value to relieve fluid retention complicating obesity.

SUPPLIED: 250 mg. and 500 mg. scored tablets 'DIURIL' (chlorothiazide); bottles of 100 and 1,000.

MSD MERCK SHARP & DOHME

Division of MERCK & CO., INC., Philadelphia 1, Pa.

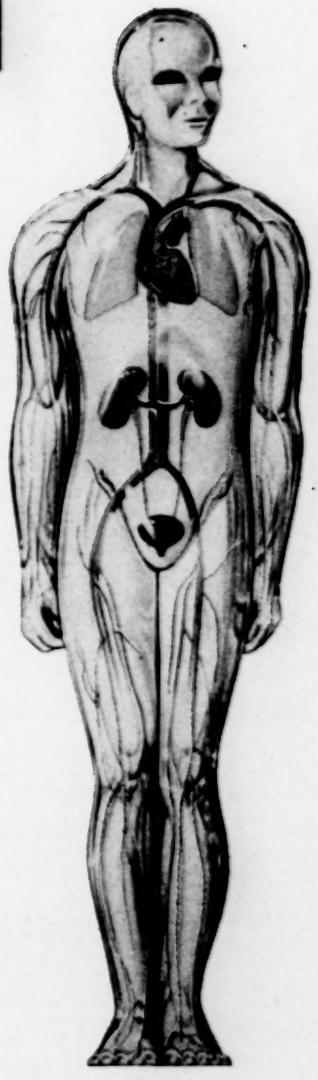
as simple
as 1-2-3
in

HYPERTENSION

1 INITIATE 'DIURIL' THERAPY
'DIURIL' is given in a dosage range of from 250 mg. twice a day to 500 mg. three times a day.

2 ADJUST DOSAGE OF OTHER AGENTS
The dosage of other antihypertensive medication (reserpine, veratrum, hydralazine, etc.) is adjusted as indicated by patient response. If the patient is established on a ganglionic blocking agent (e.g., 'INVERSINE') this should be continued, but the total daily dose should be *immediately* reduced by 25 to 50 per cent. This will reduce the serious side effects often observed with ganglionic blockade.

3 ADJUST DOSAGE OF ALL MEDICATION
The patient must be frequently observed and careful adjustment of all agents should be made to determine optimal maintenance dosage.



BENEFITS:

- **improves and simplifies the management of hypertension**
- **markedly enhances the effects of antihypertensive agents**
- **reduces dosage requirements for other antihypertensive agents—often below the level of distressing side effects**
- **smooths out blood pressure fluctuations**

INDICATIONS: management of hypertension

Smooth, more trouble-free management of hypertension with 'DIURIL'

next time, try...

PARACORT*
PREDNISONE
PARKE-DAVIS

or

PARACORTOL*
PREDNISOLONE
PARKE-DAVIS

THREE TO FIVE TIMES THE ACTIVITY OF CORTISONE OR HYDROCORTISONE

supplied

PARKE, DAVIS & COMPANY · DETROIT 32, MICHIGAN

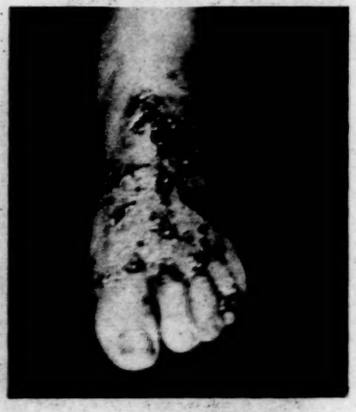
outstanding efficacy in **skin disorders**

STEROSAN®
Hydrocortisone

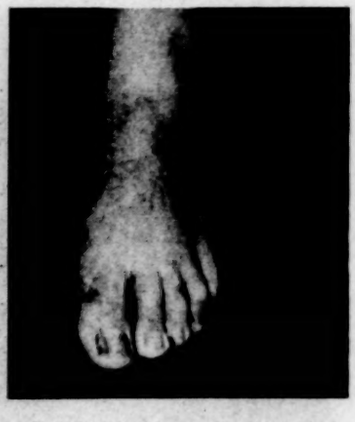
Cream and Ointment (chlorquinaldol GEIGY with hydrocortisone)

The case illustrated below typifies the superior response produced by STEROSAN-Hydrocortisone. Combining potent antibacterial-antifungal action with a reliable anti-inflammatory and antipruritic effect, STEROSAN-Hydrocortisone is valuable in a wider range of infective or allergic dermatoses.

A severe infectious eczematoid dermatitis on foot of 15-year-old boy. Patient used STEROSAN-Hydrocortisone preparation 3 times a day for 23 days with a dramatic improvement as shown.*



before treatment



after treatment

*Case report and photographs through the courtesy of N. Orentreich, M.D., New York, N.Y.

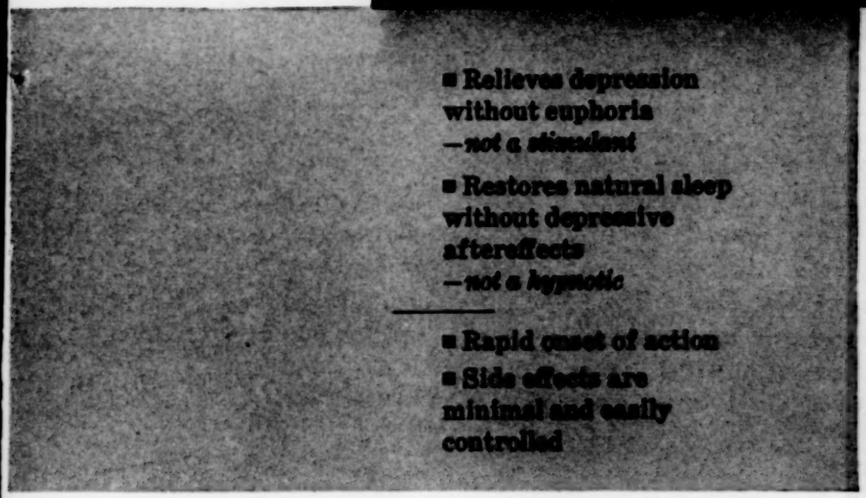
STEROSAN®-Hydrocortisone (3% chlorquinaldol GEIGY with 1% hydrocortisone) Cream and Ointment. Tubes of 5 Gm. Prescription only.

GEIGY ARDSLEY, NEW YORK

89558

NOW

COUNTERACT DEPRESSED MOODS *without stimulation*



- Relieves depression without euphoria —*not a stimulant*
- Restores natural sleep without depressive aftereffects —*not a hypnotic*

- Rapid onset of action
- Side effects are minimal and easily controlled

▲Deprol▲

Composition: Each tablet contains 400 mg. meprobamate and 1 mg. benactyzine HCl

Average Adult Dose: 1 tablet q.i.d.

WALLACE LABORATORIES, *New Brunswick, N. J.* ***Literature and samples on request***

CD-6187

A new concept in antihypertensive therapy: concomitant use of an improved ganglionic blocking agent ('Inversine') and a new antihypertensive agent ('Diuril') for smoother, simplified management of hypertension.

Longer Life for Hypertensives

In moderate, severe, and malignant hypertension, ganglionic blocking 'Inversine' often makes possible a lessening of cardiovascular-renal damage, regression of the basic disease, and prolongation of life.

"When employed under carefully controlled conditions with adequate attention to proper regulation of dosage, mecamylamine ['Inversine'] may be expected to reduce blood pressure effectively and to ameliorate various manifestations of hypertensive-cardiovascular disease. These include such symptoms as headache, dizziness and vertigo, hypertensive encephalopathy and cerebral or subarachnoid hemorrhage, retinopathy, cardiac hypertrophy, and, in some cases, cardiac decompensation."

Council on Pharmacy and Chemistry, New and Nonofficial Remedies: Mecamylamine Hydrochloride, J.A.M.A. 162: 1469-1471, Dec. 15, 1956.

Now, concomitant use of a newly discovered antihypertensive agent ('Diuril') has been found to enhance the hypotensive effect of 'Inversine'—while reducing the required dosage of 'Inversine' and often minimizing the serious side effects of ganglionic blockade.

'Inversine'
MECAMYLAMINE HYDROCHLORIDE

a greatly improved ganglionic blocking agent

Unlike the other ganglionic blocking agents, 'Inversine' is not a quaternary ammonium compound. It is a secondary amine, and has significant advantages over all other ganglionic blocking drugs:

- of the orally effective blocking agents, only 'Inversine' is completely and uniformly absorbed
- it provides predictable, reproducible effects with minimal day-to-day fluctuations in blood pressure response
- 'Inversine' is effective in low dosage
- permits convenient dosage schedules
- usefulness not limited by development of tolerance
- it has a gradual onset of effect, reducing the likelihood of sudden drops in blood pressure

'Diuril'
CHLOROTHIAZIDE

new and unique antihypertensive agent

- provides basic therapy to improve and simplify the management of hypertension
- often reduces dosage requirement of ganglionic blocking agents and other antihypertensive agents below the level of serious side effects
- added to other antihypertensive agents, is often effective in controlling blood pressure of even highly resistant cases
- smooths out blood pressure fluctuations
- effectiveness not diminished by development of tolerance
- well tolerated even at maximum therapeutic doses

DOSAGE RECOMMENDATIONS

New Patients

1. *Initiate 'Diuril' therapy*

'Diuril' is given in a dosage range of from 250 mg. twice a day to 500 mg. three times a day, depending on severity of the hypertension.

2. *Add 'Inversine' as follows:*

('Inversine' is established in the same manner whether used with 'Diuril' or alone.) Recommended initial dosage is 2.5 mg. 'Inversine' twice a day, preferably after meals. May be increased by 2.5 mg. at intervals of no less than two days until desired response is obtained. In severe or urgent cases, the increments may have to be larger or more frequent, with the largest dose given preferably at noon or in the evening. 'Inversine' is extremely potent and should always be titrated according to the patient's orthostatic blood pressure response.

3. *Adjust dosage of 'Diuril' for optimal response.*

Patients on 'Inversine' and/or other ganglionic blocking agents

1. *Initiate 'Diuril' therapy*

'Diuril' is given in a dosage range of from 250 mg. twice a day to 500 mg. three times a day, depending on severity of the hypertension.

2. *Adjust dosage of ganglionic blocking agent*

If the patient is established on a ganglionic blocking agent (e.g., 'Inversine') it should be continued, but the total daily dosage should *immediately* be reduced by as much as 25 to 50 per cent. This will reduce the serious side effects often observed with ganglionic blockade.

If other antihypertensive agents are used, their dosage should be adjusted as indicated by patient response.

3. *Determine optimal maintenance dosage*

The patient *must* be observed frequently and careful adjustment of all agents should be made to determine optimal maintenance dosage.

PRECAUTIONS: Side effects of 'Inversine' are essentially the same as those encountered with other ganglionic blocking agents. At the first sign of constipation, vigorous treatment must be initiated immediately since paralytic ileus may result if constipation is unchecked. Patients should be informed how to cope with postural hypotension should this occur. 'Inversine' is contraindicated in coronary insufficiency, organic pyloric stenosis and recent myocardial infarction.

SUPPLIED: 'Inversine', tablets of 2.5 mg. and 10 mg. Bottles of 100. 'Diuril', tablets of 250 mg. and 500 mg. Bottles of 100 and 1000.

Inversine
MECAMYLAMINE HYDROCHLORIDE

Diuril
CHLOROTHIAZIDE

MSD **MERCK SHARP & DOHME,** DIVISION OF MERCK & CO., INC., PHILADELPHIA 1, PA.

INTENSIFIED BROAD-SPECTRUM ANTIBIOTIC CONTROL

Tetrex® CAPSULES

Tetracycline Phosphate Complex U. S. Pat. 2,791,609

often the difference between rapid and delayed response

blood levels practically double those of tetracycline hydrochloride within 1-3 hours/ *maintains higher blood levels* than tetracycline hydrochloride up to 24 hours/ *a single, highly efficient antibiotic* permitting simple, flexible dosage/ *equally effective on convenient b.i.d. schedule,* as on a q.i.d. schedule/ *practically sodium-free*— pure compound—*not a mixture.*

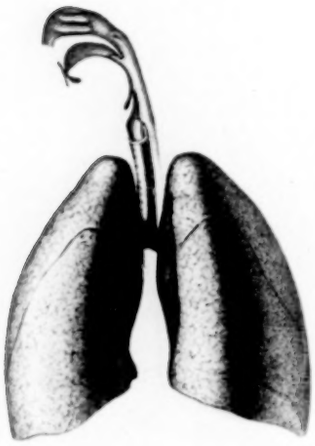
Supplied: TETREX Capsules containing the equivalent of 250 mg. tetracycline HCl activity; bottles of 16 and 100. New TETREX Pediatric Capsules containing the equivalent of 100 mg. tetracycline HCl activity; bottles of 25 and 100.

Tetrex®

Tetracycline Phosphate Complex 250 • 100 mg. CAPSULES

Bristol
LABORATORIES INC.
SYRACUSE, NEW YORK

"QUANAT" REPRODUCTION, PAT. PENDING.



Tetrex®-APC

WITH BRISTAMIN®

Bristol
LABORATORIES, INC.
SYRACUSE, NEW YORK

'QUANAT' REPRODUCTION, PAT. PENDING.

INTENSIFIED TETRACYCLINE CONTROL

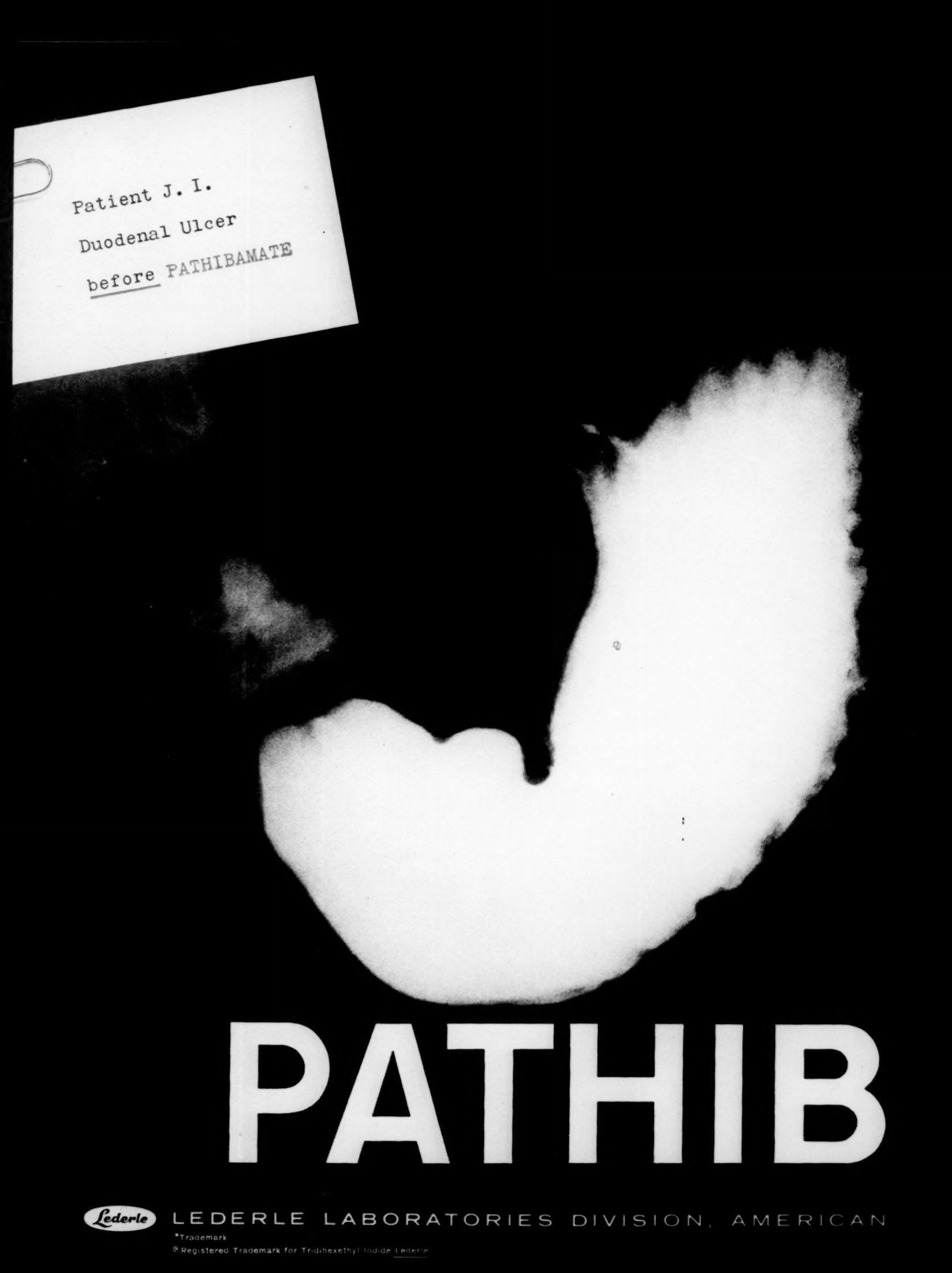
Integrated with ANALGESIA CONTROL / ANTIHISTAMINE CONTROL

new for respiratory infections

Tetrex®-APC WITH BRISTAMIN®

▪ *The only anti-infective preparation providing* "all-factor" control through the (1) faster, higher blood levels and broad-range activity of TETREX (2) analgesic-antipyretic action of APC (3) notably potent antihistamine effects of BRISTAMIN, virtually free of somnolence. ▪ *Specific therapy* for bacterial infections caused by tetracycline-sensitive organisms. *Ideal adjunctive therapy* in common upper respiratory tract infections for control of secondary bacterial invasion. ▪ *Convenient, economical.*

Each capsule contains:	TETREX (tetracycline phosphate complex) (tetracycline HCl activity)	125 mg.
	Aspirin	150 mg.
	Phenacetin	120 mg.
	Caffeine	30 mg.
	Bristamin (Phenyltoloxamine Citrate)	25 mg.
Usual dose:	One or two capsules q.i.d.	
Supplied:	Bottles of 24 and 100.	



PATHIB

Lederle LEDERLE LABORATORIES DIVISION, AMERICAN

*Trademark

® Registered Trademark for Tridihexethyl Iodide Lederle

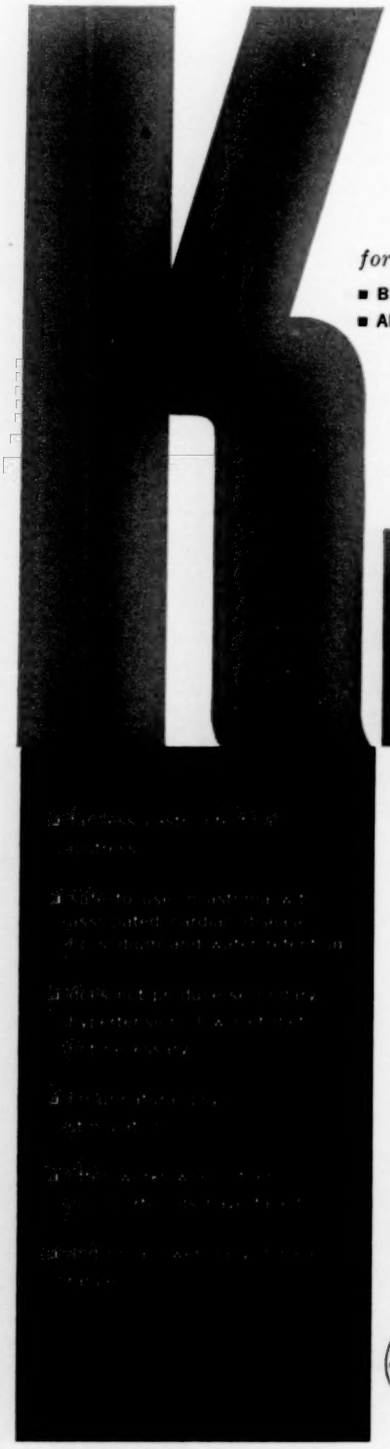
. . . calms tension and controls G. I. trauma

AMATE*

Meprobamate with PATHILON® Lederle

CYANAMID COMPANY, PEARL RIVER, NEW YORK

A NEW, CORTICOSTEROID MOLECULE WITH GREATER ANTIALLERGIC, ANTIRHEUMATIC AND ANTI-INFLAMMATORY ACTIVITY



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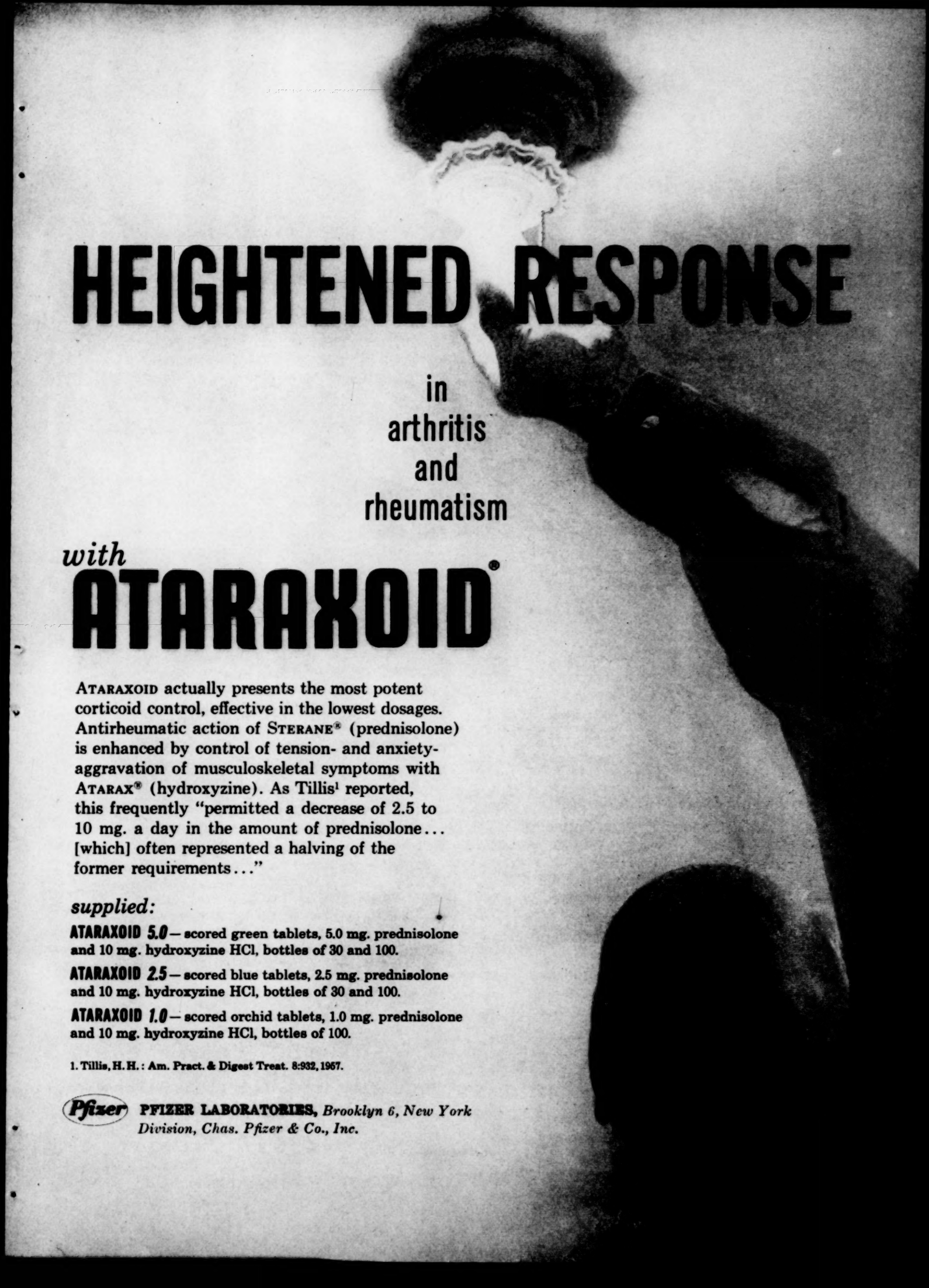

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1. Tillis, H. H.: Am. Pract. & Digest Treat. 8:932, 1957.

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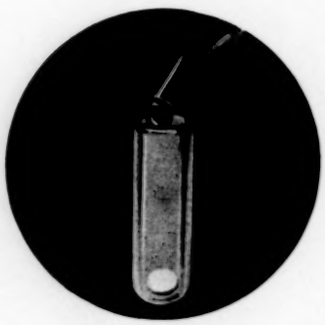

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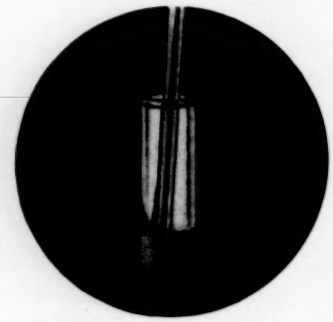

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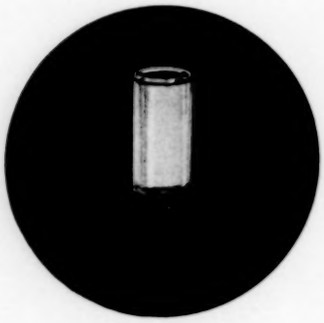

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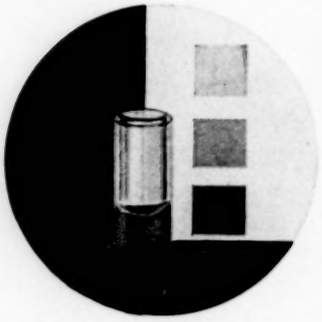

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REFERENCES—(1) Settel, E.: Personal communication. (2) Holley, H. L.: Personal communication. (3) Burns, J. J.; Trousof, N., and Brodie, B. B.: To be published. (4) Smith, R. T.: To be published. (5) Peak, W. P., and Smith, R. T.: To be published. (6) Wiesel, L. L.: Personal communication. (7) Passarelli, W. W.: Personal communication.

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Wiesel[6]	advanced osteoarthritis	12	10	less muscle spasm and pain
Passarelli[7]	degenerative and rheumatoid arthritis	9	9	improvement, with less stiffness and freer motion
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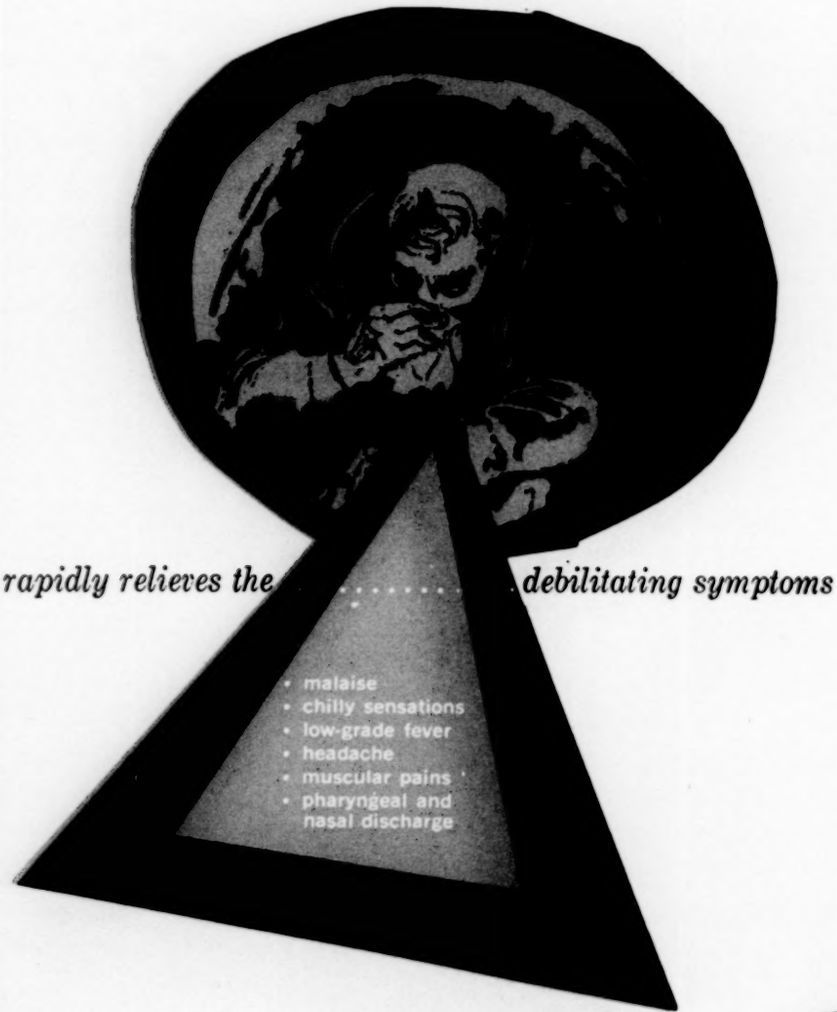

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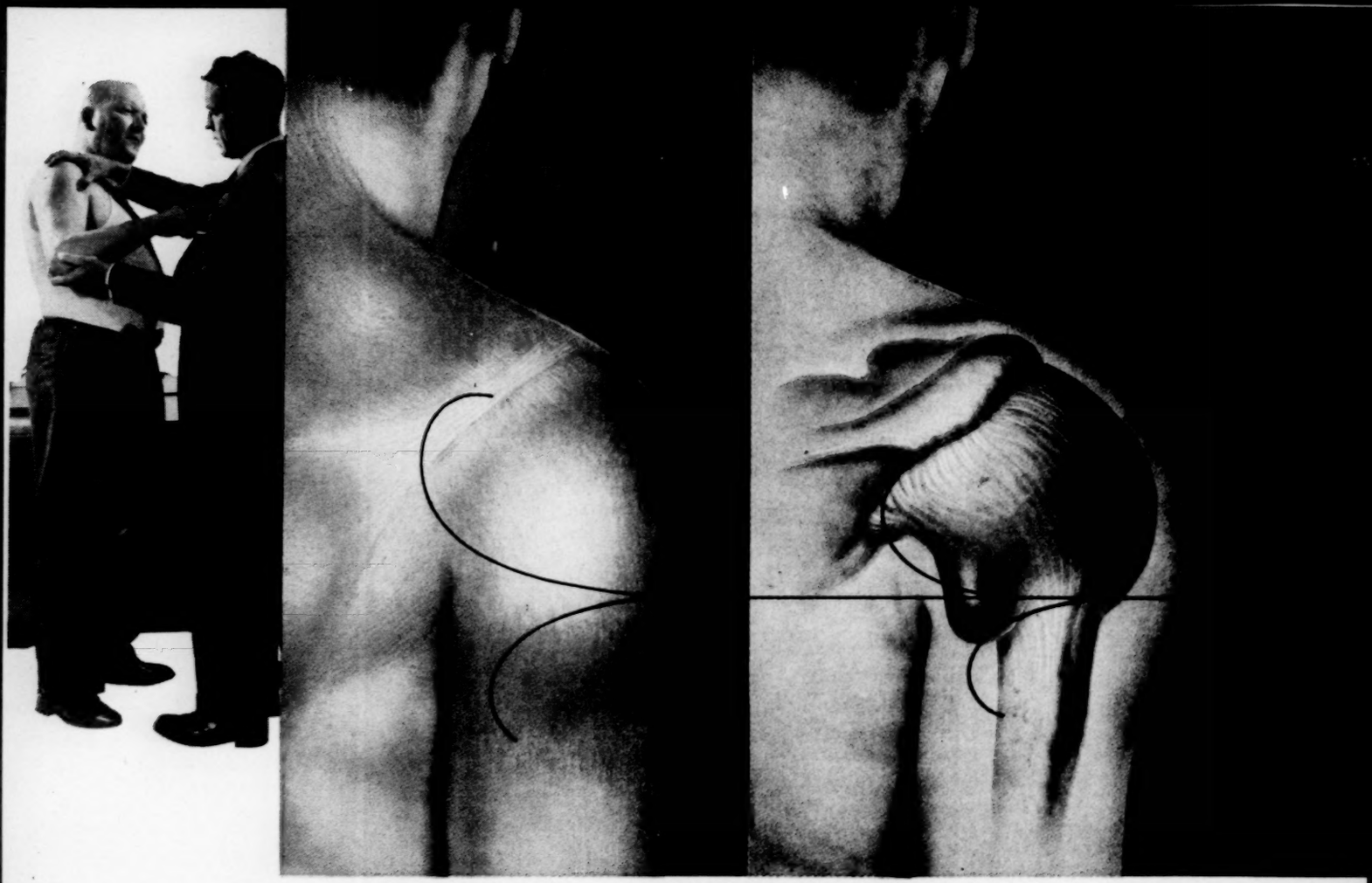

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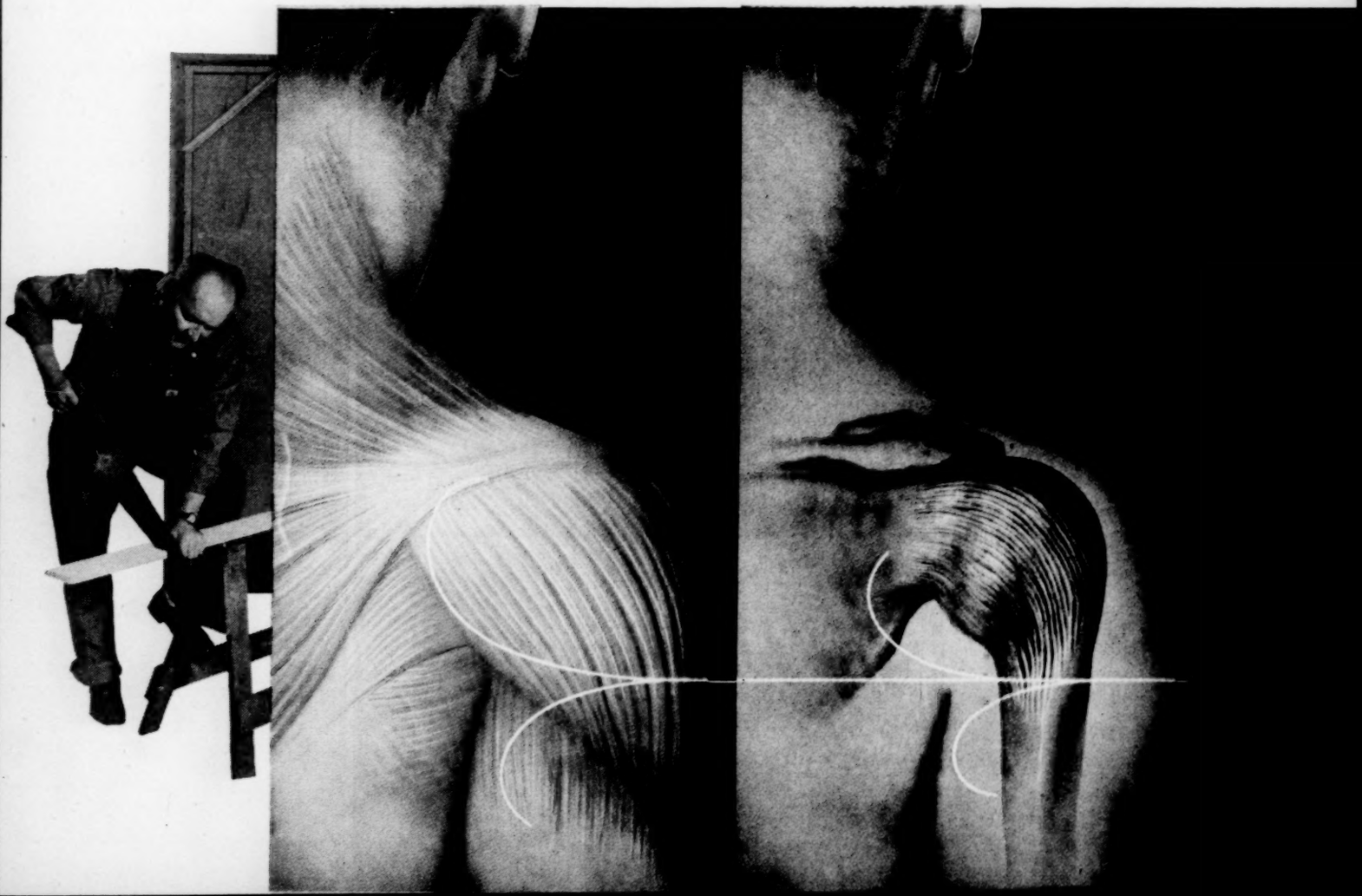

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1. Comroe's Arthritis: Hollander, J. L., p. 149 (Fifth Edition, Lea & Febiger, Philadelphia, Pa. 1953).
2. Merck Manual: Lyght, C. E., p. 1102 (Ninth Edition, Merck & Co., Inc., Rahway, N. J. 1956).

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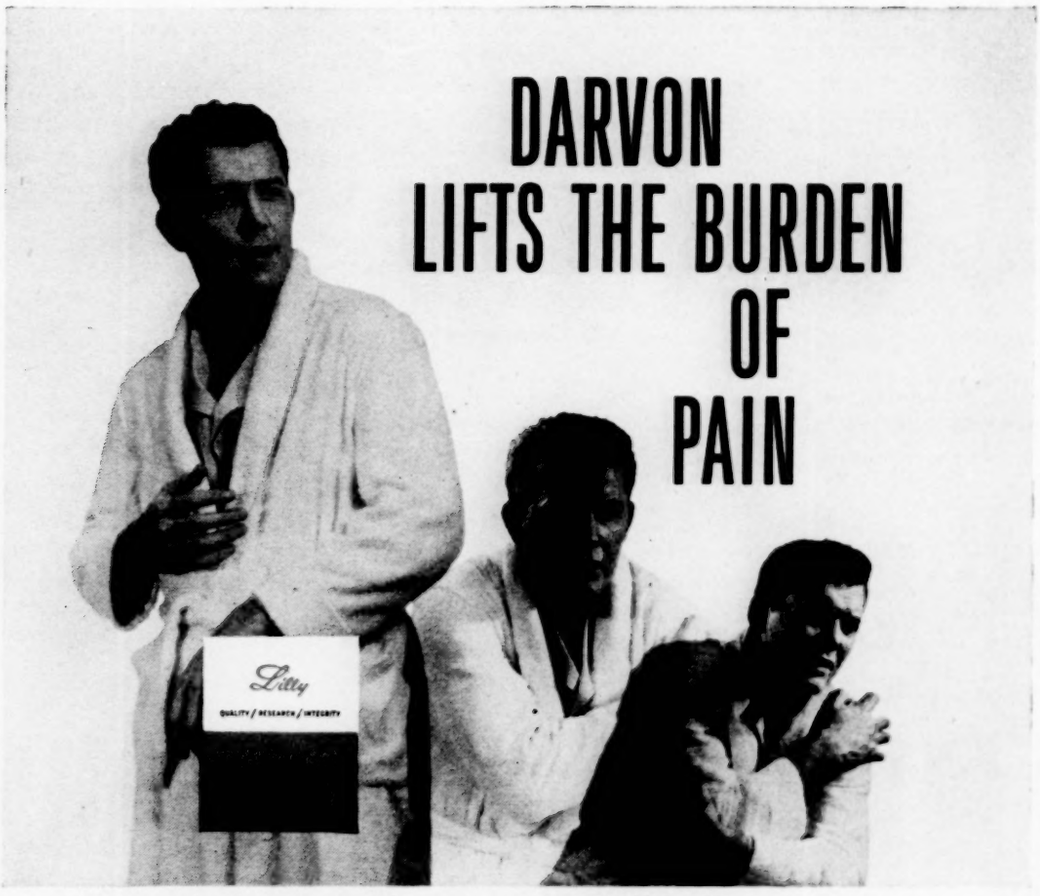

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The American Journal of Medicine

VOL. XXIV APRIL, 1958 NO. 4

Editorial

The Alveolar-Capillary Block Syndrome

IN 1951 Austrian et al. [1] defined the term "alveolar-capillary block syndrome" and called attention to its role in the causation of pulmonary insufficiency of various etiology.

Clinical Features. From the clinical point of view [1,2] the syndrome is characterized by hyperventilation, dyspnea, tachypnea, cyanosis, at first only on exercise but subsequently also at rest, basal rales, absence of wheezing or other signs of endobronchial obstruction and, late in the course of the disease, by signs and symptoms of right heart failure. Clubbing may or may not be present. A chest roentgenogram usually reveals diffuse pulmonary infiltration.

Physiological Features. From the physiological point of view [1,2] the alveolar-capillary block syndrome is characterized principally by a reduction in the diffusing capacity of the lung. Other findings include uniform reduction in lung volume, normal residual volume/total capacity ratio (unless this is associated with emphysema), well preserved maximum breathing capacity, normal distribution of inspired gases, hyperventilation at rest (and particularly after exercise), anoxemia on exercise (or even at rest in advanced cases), normal or decreased arterial carbon dioxide tension, elevation of pulmonary artery pressure, and often right ventricular and diastolic pressure and reduced pulmonary compliance.

Morphological Considerations. The designation alveolar-capillary block syndrome implies a block at the alveolar-capillary interface, with impairment of diffusion. This interface is not a simple boundary. In passing from the alveolus to the hemoglobin moiety of the red blood cells, oxygen must cross several fluid and tissue barriers. These include any fluid lining the alveolus, the alveolar membrane proper, the interstitial fluid of the alveolar wall, the capillary wall, the plasma in the pulmonary capillary, the red cell membrane, and the intracellular fluid of the red blood cell. The components of these structures comprising the alveolar-capillary "membrane" are commonly considered as a unit. The estimated total surface area of this membrane is extremely large, viz. 90 square meters [3].

The term alveolar-capillary block implies that, morphologically, there is involvement of the septum by some process, inflammatory, granulomatous or neoplastic. This is indeed usually the case, and reduction in diffusing capacity can commonly be attributed principally to a thickened membrane.

It must be emphasized, however, that *functionally* the surface for gas exchange consists of the total surface of ventilated alveoli which are perfused by pulmonary capillary blood. It is obvious that diffusion cannot occur either in

[1] AUSTRIAN, R., MCCLEMENT, J., RENZETTI, A., DONALD, K., RILEY, R. L. and COURNAND, A. Clinical and physiologic features of some types of pulmonary diseases with impairment of alveolar-capillary diffusion; syndrome of "alveolar-capillary block." *Am. J. Med.*, 11: 667, 1951.

[2] COURNAND, A. The syndrome of "alveolar-capillary block." Clinical physiologic, pathologic, and therapeutic considerations. Report of Annual Meeting and Proceedings. Royal College of Physicians and Surgeons of Canada, Oct. 3–4, 1952.

[3] COMROE, J. H., JR., FORSTER, R. E., II, DUBOIS, A. B., BRISCOE, W. A. and CARLSEN, E. The Lung: Clinical Physiology and Pulmonary Function Tests. Chicago, 1955. Year Book Publishers.

areas in which pulmonary capillaries perfuse non-ventilated alveoli or in ventilated alveoli which are not perfused.

Pathological changes produce reduction in the diffusing capacity not only by alteration of the thickness and physicochemical properties of the membrane but also by their effect on the surface area and pulmonary capillary blood flow. Furthermore, factors other than "block" at the alveolar-capillary interface play an important role in decreasing the diffusing capacity in pathological states not characterized by alveolar-capillary block.

Diffusing Capacity of the Lung. The diffusing capacity of the lung [*4–6*] is defined in terms of the quantity of a given gas crossing the alveolar capillary membrane per minute per mm. of pressure difference between the mean alveolar tension and the mean pulmonary capillary tension of the gas in question. For purposes of comparison, consider an *in vitro* system consisting of a gas, e.g. oxygen, contained in a closed space, separated from another closed compartment by a membrane through which the gas can diffuse. The quantity of gas diffusing across the membrane will depend upon many factors including the difference in the partial pressure of the gas on the two sides of the membrane, the thickness of the membrane as well as its physicochemical properties, the solubility of the gas in the membrane and, finally, on the surface area available for diffusion. This relatively simple system differs from the lung in two ways. First, unlike the *in vitro* system, the surface area available for diffusion is not measurable in the case of the lung; therefore, the diffusing capacity is defined for the lung as a whole. Secondly, in the *in vitro* system the gas on the distal side of the membrane is changing in concentration but not leaving the area of diffusion. In the lung, the pulmonary capillary blood flow plays an important role. It influences not only the rate of removal of the gas already diffused but also the surface area available for diffusion.

Thus the factors influencing the amount of oxygen diffused are the tension of gas on the proximal side of the alveolar capillary, the mean tension on the distal side, the time available for diffusion, the surface area and thickness of the membrane available for diffusion, the solubility of the gas, the physicochemical properties of the diffusing membranes, and finally the blood flow in the lung. An analogy, originally described by Barcroft [*7*], may be offered to clarify these relationships. Barcroft compares the diffusing process in the lung to the number of people entering a subway station and leaving on trains for some destination. This number will be influenced by the total population on the platform proximal to the turnstiles (alveolar tension of gas) and the difference in population density on the two sides of the turnstiles (pressure gradient). Depending on the size of the people, they will squeeze through the turnstiles at a greater or lesser rate of speed (solubility of the gas in the membrane). This is further influenced by the number of turnstiles (the area for diffusion) and by the speed with which they function (the properties of the membrane). Finally, the diffusion would cease altogether unless trains come along to carry the population already through the turnstiles (pulmonary capillary O_2 tension) away to their destination. This is influenced in turn by the number, the size and the frequency of these trains, and the space available in them (all this may be combined under specific hemoglobin flow).

At rest, the diffusing capacity of the lung for oxygen is normally greater than 15 cc. of oxygen per mm. pressure difference, per minute. This figure was arrived at from studies [*5,6*] using mathematical analyses of data derived from observations made during inspiration of two different levels of oxygen tension. In methods employing carbon monoxide [*4,8–10*], which has the advantage of such affinity for hemoglobin as to have a negligible plasma tension in the pul-

[4] Krogh, M. Diffusion of gases through the lungs of man. *J. Physiol.*, 49: 300, 1914.

[5] Riley, R. L., and Cournand, A. Analysis of factors affecting partial pressures of O_2 and CO_2 in gas and blood of the lungs: theory. *J. Appl. Physiol.*, 4: 77, 1951.

[6] Riley, R. L., Cournand, A. and Donald, K. Analysis of factors affecting partial pressures of O_2 and CO_2 in gas and blood of lungs: methods. *J. Appl. Physiol.*, 4: 102, 1951.

[7] Barcroft, J. Features in the Architecture of Physiological Function. Cambridge, 1934. Cambridge University Press.

[8] Forster, R. E., Fowler, W. S., Bates, D. V. and Van Lingen, B. The absorption of CO by the lungs during breath-holding. *J. Clin. Invest.*, 33: 1135, 1954.

[9] Filley, G. F., MacIntosh, D. J. and Wright, G. W. Carbon monoxide uptake and pulmonary diffusing capacity in normal subjects at rest and during exercise. *J. Clin. Invest.*, 33: 530, 1954.

[10] Marks, A., Cugell, A., Cadigan, J. and Gaensler, E. Clinical determination of the diffusing capacity of the lungs. Comparison of methods in normal subjects and patients with the "alveolar-capillary block syndrome." *Am. J. Med.*, 22: 51, 1957.

monary capillary, similar values have been found (if one converts the diffusing capacity for carbon monoxide to that of oxygen by multiplying by 1.23).

On exercise, the diffusing capacity increases [*11*], largely due to the increase in the number of pulmonary capillaries which become patent, as well as to an increase in the surface area for diffusion resulting from dilatation of the capillary bed as a whole. Under these circumstances, the maximum diffusing capacity is about four times that at rest.

The maximum diffusing capacity decreases with age, presumably due to reduction in the number and extensibility of pulmonary capillaries. In pathological states, reduction in diffusing capacity may be demonstrable solely on exercise. Such reduction in maximum diffusing capacity is due to decrease in the number of capillaries, or restriction in their ability to dilate due to involvement by disease. Not only may any increase in pulmonary blood flow be limited by disturbance in the pulmonary vascular bed but also by myocardial insufficiency due to cor pulmonale or mitral stenosis. It is obvious, therefore, that a reduction in diffusing capacity may be a consequence not only of block at the diffusing surface but also of reduction in total diffusing surface and, as a corollary, of reduction in blood flow to the lung.

Lesions involving the alveolar capillary interface, e.g., sarcoidosis, berylliosis, interstitial fibrosis, primarily result in a defect of the first type, although reduction in blood flow and area for diffusion are concomitant factors. Even such defects, however, may be physiologically measurable only on exercise (i.e., a reduction in maximum diffusing capacity). Reduction in total diffusing area may occur also when the architecture of the lung is distorted by processes other than interstitial lesions. In emphysema, primarily a ventilatory disturbance, it is not uncommon to have measurable reductions in the diffusing capacity of the lung for oxygen, even at rest. Pneumonectomy also is associated with reduced diffusing capacity due to reduction in total diffusing surface.

Differential Diagnosis. As stated previously, the alveolar-capillary block syndrome is essentially a concept, which serves not only to clarify the pathophysiology but also the diagnostic differentiation of certain pulmonary disorders. Among the diseases having the common features of the syndrome are Boeck's sarcoid, berylliosis, scleroderma of the lung, acute miliary tuberculosis, lymphangitic carcinosis [*1,2,10*] mitral stenosis [*12*], histiocytosis of the lung [*10,13*], interstitial fibrosis or granulomatosis of unknown etiology [*1,2,10*], asbestosis [*14*], eosinophilic pneumonia [*15*] and post-radiation fibrosis [*16*]. In most cases the diagnosis can be established by the associated clinical features, e.g., the skin lesions of sarcoid or scleroderma, or a history of industrial exposure. Too often, however, diagnosis hinges on lung biopsy but this too may leave the diagnosis in question.

Therapy. The syndrome further serves to provide a guide in therapy. In the absence of associated emphysema, a not infrequent accompaniment, there is usually no significant ventilatory disturbance, and secondary infection, e.g., bronchitis, is much less common. Thus use of bronchodilators usually offers no relief of respiratory distress, and antibiotics are much less frequently required. Since the primary defect is usually interstitial, there is a marked decrease in pulmonary compliance and an increase in the mechanical work and total work of breathing. Therapy must be directed at the primary lesion accounting for the alveolar-capillary block and increased rigidity of the lung. Unless the disease is infectious, e.g., tuberculosis, the therapeutic agents principally employed are the adrenal steroids, and some, albeit limited success has been achieved by their use.

In contrast to diseases of alveolar hypoventilation, e.g., emphysema, wherein carbon dioxide narcosis is a threat, particularly during oxygen therapy, no such danger exists in this category of pulmonary insufficiency. As noted previously, arterial carbon dioxide tension is

[11] Riley, R. L., Shepard, R. H., Cohn, J. E., Carroll, D. G. and Armstrong, B. W. Maximal diffusing capacity of the lungs. *J. Appl. Physiol.*, 6: 573, 1954.

[12] Carroll, D., Cohn, J. E. and Riley, R. L. Pulmonary function in mitral disease: distribution and diffusion characteristics in resting patients. *J. Clin. Invest.*, 32: 510, 1953.

[13] Renzetti, A., Eastman, G. and Auchincloss, J. H. Chronic disseminated histiocytosis X. (Schüller-Christian disease) with pulmonary involvement and impairment of alveolar-capillary diffusion. *Am. J. Med.*, 22: 834, 1957.

[14] Bader, M. E., Bader, R. A. and Selikoff, I. Pulmonary function in asbestosis of the lungs: an alveolar-capillary block syndrome. *J. Clin. Invest.*, 36: 871, 1957.

[15] Eldridge, F. Pulmonary infiltration with eosinophilia and the alveolar-capillary block syndrome. *Am. J. Med.* (In press.)

[16] Stone, D. J., Schwartz, and Green, R. A. Fatal pulmonary insufficiency due to radiation effect upon the lung. *Am. J. Med.*, 21: 211, 1956.

normal or even reduced in the alveolar-capillary block syndrome. The reason for this lies in the fact that carbon dioxide is about twenty times more diffusable than oxygen, and even in advanced cases of interstitial involvement there is little difficulty in passage of carbon dioxide across the membrane; indeed if there were the condition would be incompatible with life unless continuous oxygen therapy were administered. Values for arterial carbon dioxide are therefore usually normal, or indeed slightly reduced due to hyperventilation which is so commonly encountered.

Not only does one approach the problem of oxygen theory differently, but also the problem of therapy of *cor pulmonale*. Since the right heart failure in this group of patients is due primarily to restriction of the pulmonary vascular bed by organic change, and to a lesser extent to the pernicious effect of anoxemia, the results of therapy are not very happy, in contrast to the group of patients with alveolar hyperventilation in whom cor pulmonale is due to a considerable extent to hypoxemia, which is reversible. In the alveolar capillary block syndrome, the hypoxemia, if present, may be readily reversed with oxygen therapy but this measure will avail little unless steroid therapy effects improvement in the constrictive disease of the pulmonary vascular bed.

It is thus apparent that from a clinical, physiological and therapeutic standpoint the alveolar-capillary block syndrome is an extremely useful categorization of many cases of pulmonary insufficiency. Indeed, it often precisely describes the site of the pathological process. Consideration of the various components of the diffusing process, however, indicates that reduction in the capacity may not always be due to a block at the alveolar capillary interface, and explains the alterations of diffusing capacity in disease states in which no evidence for alveolar-capillary block can be obtained.

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Clinical Studies

Use of Oximes in the Treatment of Intoxication by Anticholinesterase Compounds in Normal Subjects*

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INTOXICATION by anticholinesterase compounds may occur following accidental exposure to organophosphorus insecticides (e.g., parathion [*1*] and tetraethyl pyrophosphate (TEPP) [*2*]) and chemical warfare agents (e.g., sarin) [*3,4*], and after administration of excessive amounts of any anticholinesterase compound used in the management of adbominal distention, glaucoma or myasthenia gravis. These include neostigmine [*5*], bis-neostigmine (BC 40) [*6*], pyridostigmine (mestinon®) [*7*], bis-pyridostigmine or hexamarium (BC-51), ambenomium (mytelase®) [*8*], di-isopropyl fluorophosphate (DFP) [*9*], TEPP and octamethyl pyrophosphoramide (OMPA) [*10*]. The pharmacologic effects of anticholinesterase compounds are due to the inhibition of cholinesterase enzymes in the tissues, which results in cholinergic effects attributable to the accumulation of acetylcholine in the effector organs. Accumulation of acetylcholine at the ends of postganglionic cholinergic nerves to smooth and cardiac muscle and secretory glands causes muscarine-like effects (nausea, vomiting, abdominal cramps, diarrhea, sweating, increased salivary and bronchial secretion, and bradycardia); at the ends of motor nerves to skeletal muscle, nicotine-like effects (weakness and fasciculations); and, in the central nervous system, anxiety, headache and, in some instances ataxia, coma and convulsions [*3*]. The duration of these effects is shortest following exposure to quaternary ammonium anticholinesterase compounds such as neostigmine, pyridostigmine and ambenomium, which inhibit cholinesterase enzymes reversibly; longer following exposure to partly reversible organophosphorus inhibitors such as parathion, TEPP and OMPA; and longest following exposure to more or less "irreversible" inhibitors such as DFP and sarin. The administration of large doses of atropine ameliorates the muscarine-like effects of anticholinesterase compounds, and to a lesser extent the central neural effects, but has no influence on weakness due to neuromuscular block [*3*]. This block is attributable to the accumulation of acetylcholine at the motor end-plates following the arrival of each motor nerve impulse, resulting in persistent depolarization of the end-plate region [*11*]. The block appears to persist until partial restoration of cholinesterase activity in the muscle occurs spontaneously. In severe anticholinesterase intoxication death occurs as a result of paralysis of the muscles of respiration and of the pharynx and tongue, unless artificial respiration and an open airway are maintained until spontaneous recovery occurs [*3*]. There has been no clinically useful means of accelerating recovery from the neuromuscular block.

Recently Wilson and others have demonstrated that cholinesterase inhibited by organophosphorus anticholinesterase compounds may be reactivated *in vitro* by derivatives of hydroxamic acid ($R—\overset{\overset{O}{\|}}{C}—NHOH$), and to a greater

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† Kenny Foundation Scholar.

extent by a number of oximes (R—C—R, with NOH) [*12–20*]. Both groups of compounds also react with these inhibitors to inactivate them directly. The action of organophosphorus anticholinesterase agents on smooth, cardiac and skeletal muscle of experimental animals could be reversed by hydroxamic acid derivatives [*21*] and oximes [*22,23*], and their lethal effects reduced [*24–26*], although species differences were wide.

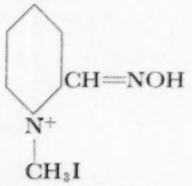
The efficacy of hydroxamic acid derivatives or oximes in reversing the action of quaternary ammonium anticholinestersae compounds, and their usefulness in man, has not been reported. This communication will present evidence that the oximes, pyridine-2-aldoxime* (2-PAM) and diacetyl monoxime* (DAM), are capable of reversing in man cholinesterase inhibition and neuromuscular block due to organophosphorus or quaternary ammonium anticholinesterase compounds, and are of clinical value in the treatment of intoxication by these compounds.

PROCEDURE

The influence of 2-PAM and DAM on the inhibition of cholinesterase enzymes of human plasma and red blood cells, and of muscle and brain (obtained postmortem) by anticholinesterase compounds *in vitro* was studied by electrometric determination of cholinesterase activity [*4,27*]. The oximes, dissolved in sodium phosphate buffer used in the enzyme assay procedure, were added to plasma, hemolyzed red cells, or tissue homogenate thirty minutes before or thirty minutes after the addition of anticholinesterase compound, and cholinesterase activity was measured over a period of one hour. The anticholinesterase compounds studied included neostigmine (prostigmin®),* pyridostigmine (mestinon®),* ambenomium (mytelase),† bis-neostigmine (BC-40),‡ bis-pyridostigmine (BC-51),‡ and sarin (isopropoxymethyl phosphonofluoridate). The effect of administration of the oximes on plasma and red blood cell cholinesterase activity of subjects in whom the activity of these enzymes had been depressed by the prior administration of each of these anticholinesterase compounds, or of OMPA,§ was also determined, employing the same method of enzyme assay.

Studies were carried out on the effect of intravascular administration of Seitz-filtered, aqueous solutions of 2-PAM and DAM on neuromuscular function, and on the neuromuscular block produced by the anticholinesterase compounds in volunteer normal subjects and convalescent patients who had no disorder of the peripheral neuromuscular system. The anticholinesterase compounds were administered intra-arterially through an indwelling 20-gauge Cournand-Riley needle in the brachial artery, as previously described [*28*], or orally. After the development of neuromuscular block and weakness, in the injected arm or generally, 2-PAM or DAM was administered intra-arterially or intravenously. Muscle function was determined by percutaneous electrical stimulation of the ulnar nerve with supramaximal pulses and recording the evoked muscle action potentials [*28*] and isometric tension [*29*] from the adductor pollicis brevis muscle. Muscle action potentials and tension produced in this way will be referred to as evoked potentials and tension. To determine the effect of drugs on the muscle response to nerve stimulation, these were injected into the brachial artery during intermittent stimulation of the ulnar nerve. The standard pattern consisted of trains of four stimuli (interval between each stimulus 40 msec.) delivered every five seconds. The injection was usually made after the fourth such train, so that a period of response to nerve stimulation was recorded as a control. The effect on the amplitude of the evoked muscle action potentials has been charted; the effect on the evoked muscle tension was similar, and has therefore been omitted from most of the

* The formula of pyridine-2-aldoxime methiodide (2-PAM) is



and of diacetyl monoxime (DAM) $CH_3—C(=O)—C(=NOH)—CH_3$

These compounds were kindly provided by Dr. Henry J. Wills, Chemical Corps Medical Laboratories, Army Chemical Center, Maryland.

* Supplied by Roche Laboratories, Nutley, New Jersey.

† Supplied by Winthrop Labs., New York City.

‡ Supplied by Merck Sharp & Dohme, Philadelphia, Pennsylvania.

§ Supplied by Eli Lilly & Co., Indianapolis, Indiana.

data. Muscle strength was also evaluated by measurement of grip strength with a hand dynamometer, and of the length of time the head or each extended leg could be elevated from the supine position, and the arms from the sitting position. Blood drawn for determination of cholinesterase activity was obtained from the brachial artery or antecubital vein of the uninjected extremity.

TABLE I
INHIBITION OF HUMAN CHOLINESTERASE ENZYMES BY 2-PAM *in vitro*

PAM Concentration (molar)	Cholinesterase Activity (ΔpH/hr.)			
	Plasma	Red Blood Cells	Muscle	Brain
0	0.33	0.24	0.61	0.52
2.6×10^{-6}	0.32	0.24	0.54	....
6.4×10^{-6}	0.35	0.24	0.58	0.47
1.3×10^{-5}	0.33	0.22	0.58	....
2.6×10^{-5}	0.32	0.22	0.56	0.45
6.4×10^{-5}	0.33	0.22	0.64	....
1.3×10^{-4}	0.31	0.21	0.60	0.42
2.6×10^{-4}	0.31	0.19	0.59	....
6.4×10^{-4}	0.27	0.16	0.57	0.33
1.3×10^{-3}	0.19	0.12	0.48	....
2.6×10^{-3}	0.16	0.10	0.42	0.17

The effect of the oximes on neuromuscular block produced by a quaternary ammonium anticholinesterase compound, neostigmine and by an organophosphorus agent, sarin, are presented in detail. Their effect on block due to the other anticholinesterase compounds studied was similar.

RESULTS

Inhibition of Cholinesterase Enzymes by Oximes in Vitro. These enzymes were inhibited by 2-PAM in concentrations above 10^{-4} M (Table I). DAM had no effect in concentrations to 10^{-2} M.

Protection and Reactivation by Oximes of Cholinesterase Enzymes Inhibited by Anticholinesterase Compounds in Vitro. (*Table* II.) 2-PAM (9 to 90 × 10^{-6} M) caused moderate protection and reactivation of human cholinesterase enzymes inhibited by sarin (2.5 to 25 × 10^{-6} M). The effect of 2-PAM on the action of the quaternary ammonium anticholinesterase agents studied was less marked, but there was slight protection of cholinesterases against inhibition by neo-

TABLE II
PER CENT PROTECTION AND REACTIVATION BY 2-PAM AND DAM OF HUMAN CHOLINESTERASE ENZYMES INHIBITED BY ANTICHOLINESTERASE COMPOUNDS *in vitro*[a]

Anticholinesterase Compound	Concentration (molar)	Per cent Protection of Cholinesterase by:								Per cent Reactivation of Cholinesterase by:							
		PAM				DAM				PAM				DAM			
		Plasma	RBC	Muscle	Brain	Plasma	RBC	Muscle	Brain	Plasma	RBC	Muscle	Brain	Plasma	RBC	Muscle	Brain
Neostigmine	6×10^{-7} to 1×10^{-5}	10	25	34	11	8	20	30	6	0	15	13	16	0	0	4	8
Pyridostigmine	3.5×10^{-7} to 3.5×10^{-5}	22	21	4	6	1	16	0	15	14	8	6	0	0	5	0	8
Ambenonium	3×10^{-6} to 3×10^{-5}	6	..	..	..	11	..	..	..	6	..	..	..	7	..	..	..
	1.5×10^{-5} to 1.5×10^{-4}	..	1	0	5	..	0	0	0	..	3	0	14	..	0	0	0
Sarin	2.5×10^{-6} to 2.5×10^{-5}	20	30	50	54	5	13	16	19	25	30	63	57	0	2	0	

[a] 2-PAM (9×10^{-6} to 9×10^{-5} M) or DAM (9×10^{-6} to 2×10^{-3}) was added to the enzyme preparation thirty minutes before (for protection) or thirty minutes after (for reactivation) addition of anticholinesterase compound, and cholinesterase activity then measured after one hour at 24°C. Maximal values are recorded. The cholinesterase activity in the absence of inhibitor or oxime was 0.6 to 0.8 ΔpH/hour.

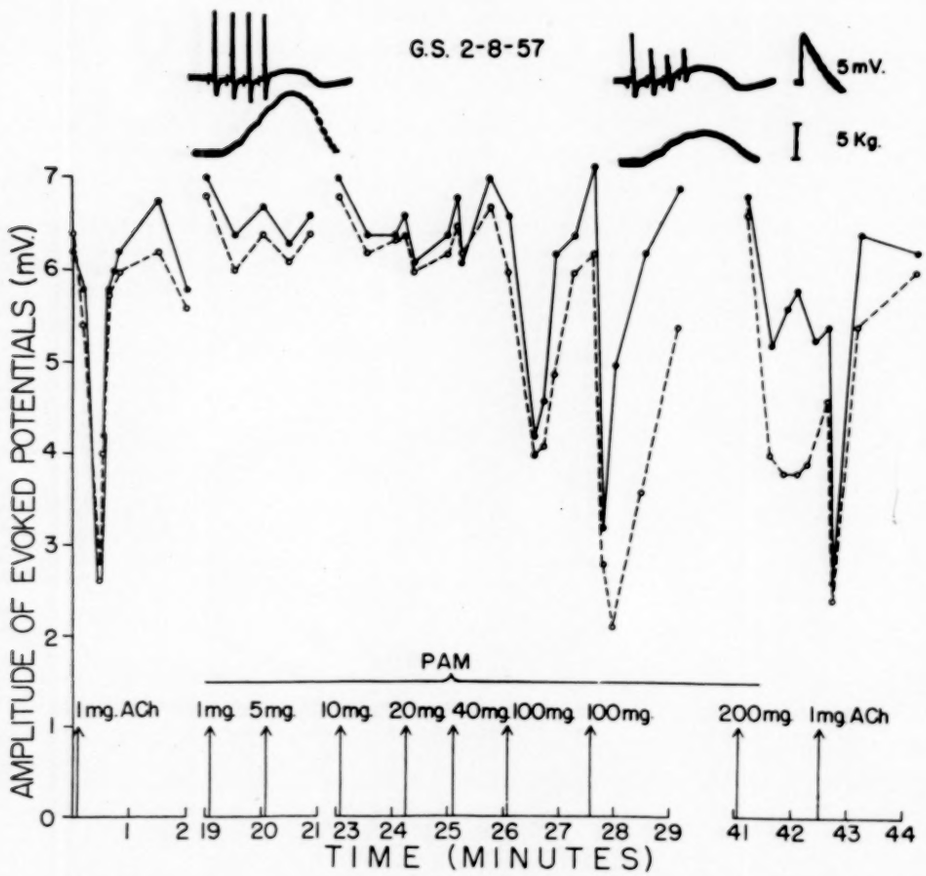


FIG. 1. Depression of evoked muscle action potentials and tension produced by large doses of 2-PAM, and lack of effect of this block on the prompt depressant action of acetylcholine. The amplitude of the first (●——●) and fourth (○——○) muscle action potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted, and illustrative potentials (mV.) and tension (kg.) recorded above. Injections were intra-arterial.

stigmine and pyridostigmine (10^{-7} to 10^{-5} M), and very slight reactivation of the enzymes following inhibition.

DAM (9×10^{-6} to 2×10^{-3} M) was less effective than 2-PAM, but did protect cholinesterases to a slight degree against inhibition by sarin, neostigmine, and possibly pyridostigmine. In contrast to 2-PAM, DAM did not reactivate the enzymes following inhibition, with the possible exception of muscle cholinesterase inhibited by neostigmine. Neither appreciably protected or reactivated cholinesterases inhibited by ambenomium (10^{-9} to 10^{-5} M).

Effect of Administration of Oximes. Intravenous administration: 2-PAM and DAM were administered on fifteen occasions to seven subjects in doses of 500 to 2,000 mg. The injection of 2-PAM at a rate of 100 to 300 mg./minute produced no signs or symptoms, no change in the blood pressure, recumbent or standing, and no change in the cardiac rate, recumbent. On standing, three of the subjects had an increase in cardiac rate from a mean of 80 to 105 per minute; this increase had not occurred on standing prior to the administration of 2-PAM.

The injection of DAM at the rate of 60 to 300 mg./minute produced a burning sensation at the site of injection radiating up the injected vein to the shoulder, followed by moderate giddiness, drowsiness and a sensation of warmth and tingling in the abdomen and chest. These symptoms lasted from one to five minutes after cessation of the injection. In three subjects there was no change in the blood pressure, recumbent; in one it increased from 110/64 to 134/84 mm. Hg, and in three it decreased slightly, from a mean of 128/75 to 108/72 mm. Hg. In the latter three subjects, and in one in whom there was no

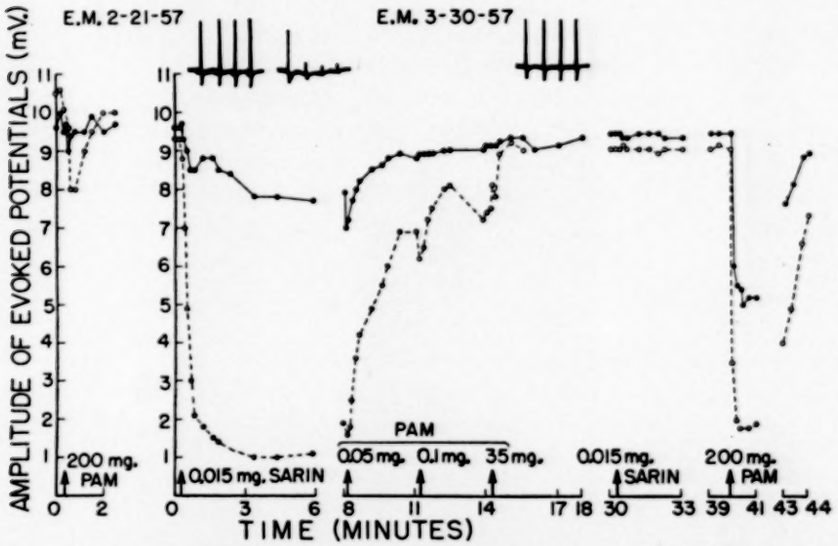


FIG. 2. Reversal by 2-PAM of the depressant effect of sarin on evoked muscle action potentials; inhibition by PAM of the depressant effect of a second injection of sarin; and increase in the depressant effect of PAM following the administration of sarin (right, control on left). The amplitude of the first (●———●) and fourth (○———○) muscle action potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted, and illustrative potentials recorded above. Injections were intra-arterial.

change in the blood pressure, recumbent, there was a moderate reduction in the blood pressure on standing from a mean of 126/84 mm. Hg prior to DAM to 103/68 mm. Hg after DAM. In only one subject was there an increase in cardiac rate when recumbent, from 84 to 104 per minute, but all who had a reduction in blood pressure on standing had an accompanying increase in cardiac rate, from a mean of 90 to 105 per minute. The injection of DAM at the rate of 40 mg./minute resulted in no local or systemic symptoms.

Neither 2-PAM nor DAM in the doses administered produced any alteration in muscle strength, plasma or red blood cell cholinesterase activity, blood counts, urinalysis, hepatic or renal function, or in the electrocardiogram.

Intra-arterial Administration of 2-PAM. Neuromuscular block produced by large doses: 2-PAM was administered in doses up to 800 mg. The injection of 40 mg. resulted in a slight burning sensation in the injected extremity, and 100 mg. in moderate burning. Five to 40 mg. produced 5 to 10 per cent reduction in the amplitude of evoked muscle action potentials lasting less than a minute, while 100 to 200 mg. produced approximately 50 per cent reduction lasting one to three minutes. (Fig. 1.) The effect of successive injections was cumulative only when administered at intervals of less than four minutes. An initial injection of 100 mg. produced even depression of successive potentials evoked by a train of four stimuli at 40 msec. intervals, while a second injection of 100 mg. two minutes later produced progressive depression (decrement) of successive evoked potentials. Owing to the transient nature of the block, its properties were difficult to study. However, there appeared to be no effect of the block on the prompt depressant action of intra-arterially injected acetylcholine. (Fig. 1.) The block was not reversed by either acetylcholine or neostigmine, nor was the rate of recovery from the block affected by these drugs. Following the injection of anticholinesterase compounds there was a moderate increase in the depressant effect of 2-PAM on evoked potentials, and an increase in the decrement of these potentials. (Fig. 2.) Following the injection of 2-PAM there was a decrease in the depressant effect of anticholinesterase compounds on evoked potentials. (Figs. 2 and 3.)

Intra-arterial Administration of DAM. Lack of effect on neuromuscular transmission: DAM was administered in doses up to 400 mg., without effect on neuromuscular transmission. The prompt depressant action of intra-arterially injected acetylcholine was also unchanged. Twenty mg. of DAM produced moderate burn-

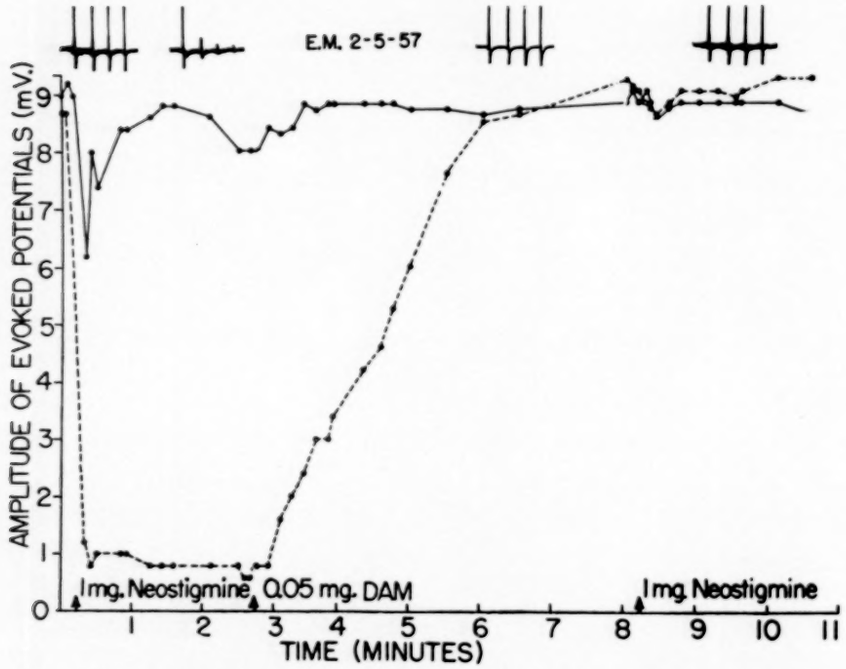


FIG. 3. Reversal by DAM of the depressant action of neostigmine on evoked muscle action potentials, and inhibition by DAM of the depressant effect of a second injection of neostigmine. The amplitude of the first (●———●) and fourth (○———○) potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted, and illustrative potentials recorded above. Injections were intra-arterial.

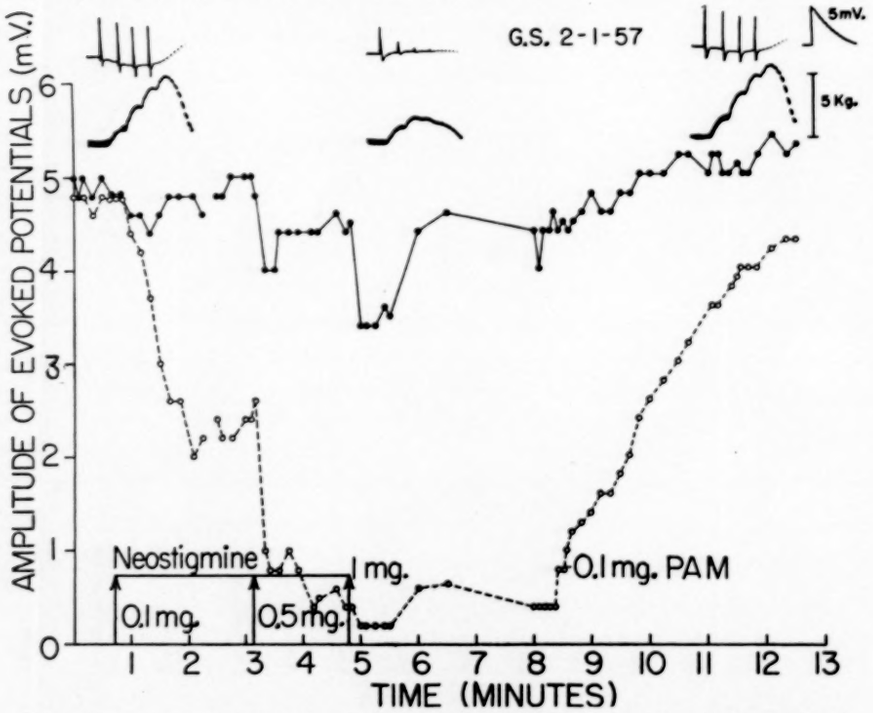


FIG. 4. Reversal by 2-PAM of the depressant effect of neostigmine on evoked muscle action potentials and tension. The amplitude of the first (●———●) and fourth (○———○) potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted, and illustrative potentials (mV.) and tension (kg.) recorded above.

ing, and 40 mg. severe burning and moderate redness of the injected extremity. The systemic effects of DAM and of 2-PAM were the same as following intravenous administration.

Effect of Administration of Oximes on the Actions of Anticholinesterase Compounds. Reversal by intra-arterial 2-PAM and DAM of neuromuscular block produced by anticholinesterase compounds: Neuromuscular block was produced by the intra-arterial injection of each of the following anticholinesterase compounds: neostigmine, bis-neostigmine, pyridostigmine, bis-pyridostigmine, ambenomium and sarin. There was progressive depression of successive evoked muscle action potentials and repetitive firing after the initial potential of a train. (Figs. 2–6.) Fasciculations occurred in the injected extremity and, in most instances, generally. Following the doses administered, the latter potentials of the train remained markedly depressed for ten to thirty minutes and then gradually returned to normal over a period of twenty to sixty minutes. (Figs. 5 and 6.) The neuromuscular block produced by each of the anticholinesterase compounds was promptly and strikingly reversed in the injected extremity immediately after the intra-arterial injection of 0.05 to 0.1 mg. of 2-PAM or DAM. (Figs. 2, 3 and 4.) There was also reduction or disappearance of repetitive firing and fasciculations in the injected extremity. Five to ten seconds after the injection of oxime the depressed potentials began to increase, and within one to three minutes after injection they approached the initial amplitude. 2-PAM and DAM were approximately equally effective, and the action of each of the anticholinesterase compounds was influenced to the same degree. When the oximes were injected simultaneously or in sequence their effect was additive. Following either partial or complete reversal of neuromuscular block due to quaternary ammonium anticholinesterase compounds, there was no subsequent decline in amplitude of the evoked potentials; reversal of the block was permanent. (Fig. 3.) Following reversal of neuromuscular block due to sarin, there was in some instances a slight decline in amplitude of the potentials. (Fig. 2.)

Following restoration of the potentials to their initial amplitude, the subsequent injection of

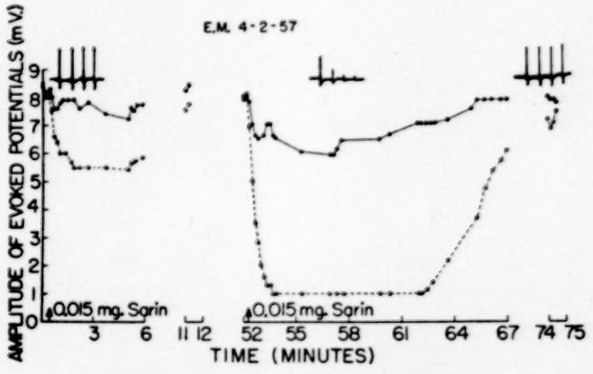


FIG. 5. Time course of the depressant effect of sarin on evoked muscle action potentials. The amplitude of the first (●——●) and fourth (○——○) potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted, and illustrative potentials recorded above.

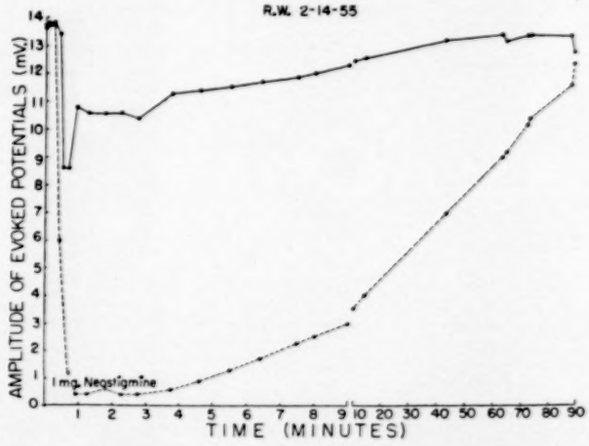


FIG. 6. Time course of the depressant effect of neostigmine on evoked muscle action potentials. The amplitude of the first (●——●) and fourth (○——○) potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted. Injection was intra-arterial.

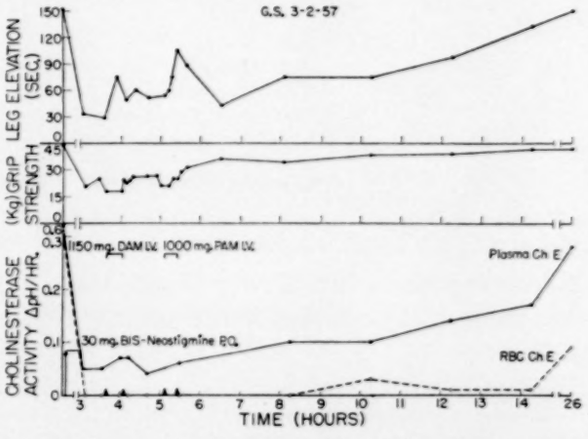


FIG. 7. Reversal by DAM and 2-PAM of generalized weakness produced by bis-neostigmine, and transient reversal of plasma cholinesterase inhibition.

TABLE III

EFFECT OF ADMINISTRATION OF 2-PAM AND DAM ON THE CHANGES IN GENERAL STRENGTH AND CHOLINESTERASE ACTIVITY (PLASMA AND RED BLOOD CELL) PRODUCED BY ANTICHOLINESTERASE COMPOUNDS

Patient	Anticholinesterase Compound	Dose (mg.)	Route*	Effect of Anticholinesterase Compound:			Oxime i.v.† (mg.)	Effect of Oxime		Cholinesterase Activity (ΔpH/hr.)					
										Plasma			Red Blood Cell		
				Gastrointestinal Symptoms	Weakness	Fasciculation		Increase in Strength	Decrease in Fasciculation	Control	Before Oxime	Increase After Oxime	Control	Before Oxime	Increase After Oxime
PAM															
W. F.	Neostigmine	1.5	i.a.	+	0	++	200	....	0	0.52	0.26	0.05	0.73	0.65	0
G. S.	Neostigmine	2.5	i.a.	+	+	++	800	+	±	....	....	....	....	....	....
G. S.	Bis-neostigmine	30	p.o.	++++	+++	+++	1,000	++	+	0.62	0.04	0.02	0.62	0	0
G. S.	Bis-pyridostigmine	1.3	i.a.	0	+	+	150	±	±	0.62	0.08	0.05	0.60	0.24	0.02
G. S.	Ambenomium	0.1	i.a.	++	+++	+++	700	++	+	0.65	0.28	0.07	0.59	0.22	0.04
E. M.	Ambenomium	0.17	i.a.	++	+++	+++	400	++	+	....	....	....	....	....	....
C. E.	OMPA	40	p.o.	0	0	0	1,000	....	..	0.81	0.28	0.02	0.69	0.38	0.05
C. E.	OMPA	40	p.o.	0	0	0	1,000 (48 hr. after OMPA)			0.81	0.36	0	0.69	0.46	0
E. M.	Sarin	0.07	i.a.	0	0	0	1,000	....	..	0.86	0.71	0.15	0.60	0.42	0.10
DAM															
E. M.	Neostigmine	4.5	i.a.	++	+	+++	200	±	±	....	....	....	....	....	....
W. F.	Neostigmine	1.5	i.a.	+	0	++	100	....	0	0.52	0.26	0.05	0.79	0.73	0.05
G. S.	Bis-neostigmine	30	p.o.	+++	+++	+++	1,000	+	±	0.62	0.05	0.02	0.62	0	0
G. S.	Bis-pyridostigmine	1.3	i.a.	0	+	+	100	0	0	0.60	0.07	0.01	0.60	0.25	0
G. S.	Ambenomium	0.1	i.a.	++	+++	+++	500	++	0	0.62	0.20	0.05	0.62	0.17	0.02
E. M.	Ambenomium	0.2	i.a.	++	++	+++	150	0	0	....	....	....	....	....	....
C. E.	OMPA	40	p.o.	0	0	0	1,000	....	..	0.81	0.31	0.03	0.69	0.46	0.01
E. M.	Sarin	0.07	i.a.	0	0	0	500	....	..	0.86	0.71	0.03	0.60	0.53	0.02

NOTE: The number of + signs denotes degree of change (range + to ++++).
* i.a. = intra-arterial, p.o. = by mouth.
† Intravenously.

2-PAM in doses of 40 to 200 mg. resulted in more depression of the evoked potentials than had been produced by this agent prior to the administration of anticholinesterase compound. (Fig. 2.) DAM did not have any depressant effect before or after the administration of anticholinesterase compound.

Protection by 2-PAM and DAM against the neuromuscular blocking action of anticholinesterase compounds: Following the intra-arterial administration of 0.05 mg. or more of 2-PAM or DAM there was marked protection against the action of anticholinesterase compounds on neuromuscular transmission. (Figs. 2 and 3.) This protective effect diminished over a period of seven to fifteen minutes.

Lack of effect of 2-PAM and DAM on neuromuscular block produced by succinylcholine: Neuromuscular block produced by the intra-arterial administration of succinylcholine in doses of 0.1 to 1 mg. was not reversed by the intra-arterial injection of 2-PAM or DAM in doses up to 250 mg. The prior administration of succinylcholine had no effect on the neuromuscular blocking action of 2-PAM.

Reversal by intravenous 2-PAM and DAM of generalized weakness produced by anticholinesterase compounds: Generalized weakness resulting from the oral or parenteral administration of each of the anticholinesterase compounds studied was ameliorated to a moderate degree following the intravenous injection of 500 to 2,000 mg. of 2-PAM or DAM (Fig. 7 and Table III). Muscular fasciculations were reduced to a lesser degree; occasionally they were unchanged. The improvement began within thirty seconds after

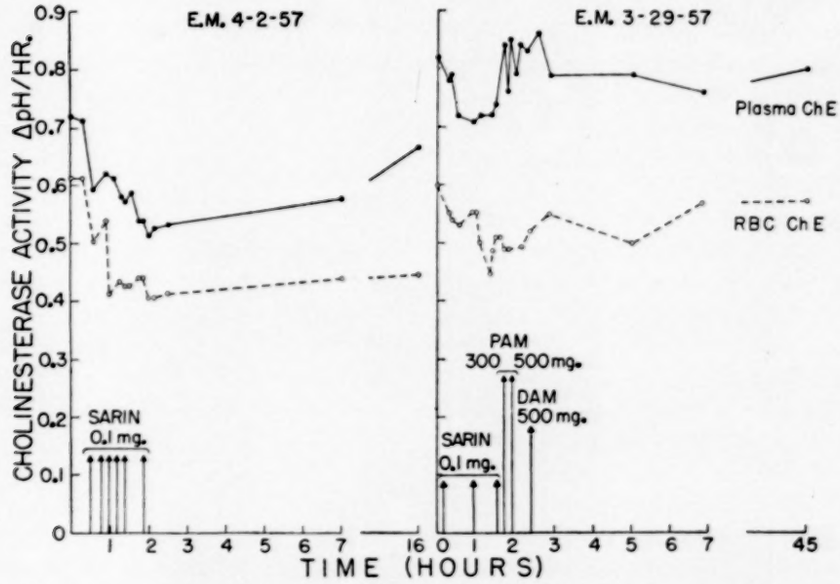


FIG. 8. Right: Reversal by 2-PAM and DAM (intravenous) of plasma and red blood cell cholinesterase inhibition produced by sarin (intra-arterial). Left: Control, effect of sarin alone.

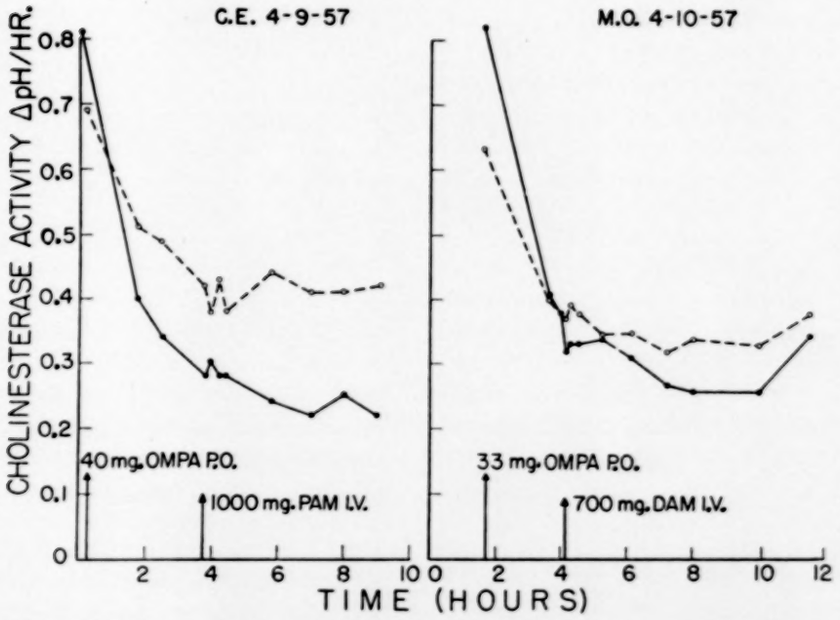


FIG. 9. Slight reversal by 2-PAM and DAM of plasma (●———●) and red blood cell (○———○) cholinesterase inhibition produced by OMPA.

injection of the oxime and was maximal in five to ten minutes. Approximately twenty minutes later there was usually some return of weakness and fasciculations, but not to the original level. (Fig. 7.) Another injection of either oxime then resulted in further improvement. The injection of 200 to 400 mg. of oxime produced slight improvement in strength in some instances, but smaller doses had no effect. The more severe the weakness and fasciculations, the less striking was the improvement following oxime administration. In no instance was there a dramatic increase in strength comparable to the reversal of neuromuscular block observed following the intra-arterial injection of much smaller doses of oxime. 2-PAM and DAM appeared to be ap-

proximately equally effective against each of the anticholinesterase compounds studied. When the oximes were administered simultaneously or in sequence their effect was additive. Neither appeared appreciably to affect the muscarine-like symptoms produced by the anticholinesterase compounds (sweating, nausea, vomiting, abdominal cramps, diarrhea and bradycardia). In contrast, the administration of atropine sulfate (1 mg., intravenously) promptly ameliorated these symptoms, but had no effect on muscular weakness or fasciculations. The effect of the oximes on central neural symptoms due to anticholinesterase compounds was more difficult to evaluate, but these did not seem to be affected as much as the muscular weakness.

Reversal by 2-PAM and DAM of plasma and red blood cell cholinesterase inhibition produced by anticholinesterase compounds: The intra-arterial or intravenous administration of 100 to 2,000 mg. of 2-PAM or DAM produced slight to moderate reversal of plasma and red blood cell cholinesterase inhibition. (Figs. 7 to 9 and Table III.) 2-PAM was somewhat more effective than DAM. The increase usually lasted fifteen to thirty minutes, and was shorter and less marked than the increase in strength. In some instances the cholinesterase activity remained elevated, although this may have been due at least in part to spontaneous restoration. The degree and duration of the restoration of cholinesterase activity varied inversely with the degree of cholinesterase inhibition, and was slight when the latter was nearly complete. (Fig. 7.) Cholinesterase inhibition produced by sarin was reversed to a greater degree and for a longer period of time by 2-PAM than that produced by the other anticholinesterase compounds, including OMPA. (Figs. 8 to 9.) This did not appear to be attributable solely to the less marked inhibition of cholinesterase produced by the doses of sarin employed.

CASE REPORT

G. S., a forty-nine year old male volunteer, was administered 30 mg. of bis-neostigmine (BC-40) orally. Two hours later he developed severe nausea, vomiting, abdominal cramps, explosive diarrhea, sweating, and slowing of the cardiac rate from 84 to 50 per minute; moderate dysarthria, dysphagia and weakness of the muscles of the neck, face and extremities; and marked generalized fasciculations. One hour later these symptoms were unchanged. Eleven hundred and fifty mg. of DAM were administered intravenously over a thirty-minute period in increments of 100 mg. injected at the rate of 4 to 20 mg./second. This resulted in a burning sensation at the site of injection (antecubital vein), radiating to the axilla. Following the injection of 500 mg. of DAM there was slight improvement in strength, but no change in fasciculations. After 1,150 mg. there was moderate improvement in strength (Fig. 7), and slight decrease in fasciculations. The blood pressure fell slightly, from 116/80 to 104/80 mm. Hg. There was no change in the gastrointestinal symptoms, sweating or bradycardia, so that the patient was then given 2 mg. of atropine sulfate intravenously, with prompt relief of these symptoms. Fifteen minutes after termination of the injection of DAM there was some return of weakness and fasciculations. The subject was then given 1,000 mg. of 2-PAM intravenously over a thirty-minute period in increments of 100 to 250 mg. injected at a rate of 4 to 20 mg./second. There were no symptoms attributable to the 2-PAM. During the half hour in which 2-PAM was injected there was a gradual and moderate improvement in strength and a slight decrease in fasciculations. Fifteen minutes after termination of the injection there was again some return of weakness, although not to the original level. One hour later the strength began to improve spontaneously and the fasciculations diminished. Strength returned to normal in about eight hours. There was slight restoration of plasma cholinesterase activity immediately after the injection of DAM and of 2-PAM, but red blood cell cholinesterase, which had been reduced to zero activity, was not affected.

COMMENTS

Reversal by Oximes of Cholinesterase Inhibition and of Anticholinesterase Neuromuscular Block. 2-PAM and DAM, which are known to reverse cholinesterase inhibition [*13–18*] and neuromuscular block [*22*] due to organophosphorus anticholinesterase compounds in experimental animals, were found in man to reverse not only the actions of these compounds but of quaternary ammonium anticholinesterase agents as well. The inhibition of cholinesterase by organophosphorus compounds has been considered to occur by direct phosphorylation of some group at the active center of the enzyme, and reversal of this inhibition by oximes to be due to displacement of the enzymic group from the phosphorus atom [*30*]. Since the quaternary ammonium anticholinesterase compounds do not contain a phosphorus atom, it is evident that the oximes may reverse cholinesterase inhibition by a more general mechanism than displacement of phosphorus.

While there were many differences between the effects of the oximes *in vitro* and *in vivo*, it

seems likely that the reversal of anticholinesterase neuromuscular block produced by these compounds is due to reactivation of muscle cholinesterase. The concentration of oxime required to reverse neuromuscular block in man was of the same order of magnitude as the concentration necessary to reverse the inhibition of human cholinesterase enzymes *in vitro*. The intra-arterial injection of 0.05 mg. of 2-PAM or DAM, if distributed throughout an estimated 200 ml. of extracellular fluid in the injected extremity would result in a concentration of 1×10^{-6} M or 2.5×10^{-6} M. Movement of the injected material down the brachial artery prior to its diffusion probably causes the local concentration in the muscles of the hand to be several fold higher. The concentration of these oximes that produced some reversal of cholinesterase inhibition *in vitro* was approximately 5 by 10^{-6} M. More direct evidence concerning the site of action of the oximes is provided by the observation of Holmes and Robins [*22*] in experimental animals that the end-plate potential, which is markedly increased in size and duration by anticholinesterase compounds, is returned to normal following the addition of oxime.

The reversal of anticholinesterase neuromuscular block by the oximes in man was more striking and more rapid than the reactivation of muscle cholinesterase by these compounds *in vitro*. Whereas these compounds had approximately the same influence *in vitro* on inhibited cholinesterase enzymes of red blood cells, brain and muscle, they were much more effective in reversing neuromuscular block due to anticholinesterase agents than in ameliorating the muscarine-like or central neural effects of these agents. These observations suggest that the enzyme in intact muscle may be particularly accessible to the action of the oximes. More difficult to explain are differences between the effects of the oximes on the action of each of the anticholinesterase compounds *in vivo* and *in vitro*. 2-PAM and DAM were equally effective in reversing anticholinesterase neuromuscular block in man, and the action of each of the quaternary ammonium and organophosphorus anticholinesterase compounds studied was influenced to the same degree. In contrast, there were marked differences in the ability of these oximes to protect against or reverse the inhibition of cholinesterase, including that of human muscle, by each of the anticholinesterase compounds *in vitro:* 2-PAM was more effective than DAM, both compounds were more effective against the organophosphorus compound studied (sarin) than against the quaternary ammonium agents, and both had little or no influence on the action of ambenomium.

The action of 2-PAM and DAM in protecting against and reversing neuromuscular block due to sarin appears to be more striking in man than in experimental animals. Holmes and Robins [*22*] found that the dose of oximes necessary to reverse neuromuscular block in the latter was close to the toxic dose; that reversal by 2-PAM could be demonstrated only after washing the muscle, since such high doses of oxime were necessary that they caused neuromuscular block; and that reversal could not be demonstrated when sarin and 2-PAM were given by intra-arterial injection. They also observed failure in the experimental animals of oximes to prevent the neuromuscular blocking action of sarin when given prior to the sarin. They attributed these results to failure of the oxime to be "fixed" in the muscle following intra-arterial injection sufficiently to maintain a high local concentration necessary to reverse or prevent anticholinesterase block. The oximes were much more effective in man following intra-arterial injection in both reversing and protecting against sarin-induced neuromuscular block.

While the oximes are capable of inactivating organophosphorus anticholinesterase compounds, including sarin, it is unlikely that this inactivation plays a part in the reversal of anticholinesterase neuromuscular block by sarin. When sarin is injected intravascularly, most of the compound reacts with esterases and other proteins within a few minutes; after an hour, it is unlikely that any uncombined sarin is present [*4*]. When the oximes were administered prior to sarin, their protective effect may have been due in part to inactivation of sarin, but the local effect of the oximes following intra-arterial injection suggests that the protection of cholinesterases from inactivation was more important.

The slow restoration of plasma and red blood cell cholinesterase activity that occurs spontaneously following inhibition of these enzymes by the administration of sarin has led to the assumption that this compound produces "more or less" irreversible inhibition of cholinesterases in man [*3,4*]. However, the neuromuscular block produced by sarin lasted only twenty to forty-five minutes, even though doses were admin-

istered which almost completely suppressed the latter potentials of a train evoked by repetitive nerve stimuli. Following comparable depression of neuromuscular transmission by DFP, which has also been regarded as a "more or less" irreversible inhibitor of cholinesterase, neuromuscular block is much more prolonged [*9*]. The duration of block produced by sarin more closely approximated that produced by TEPP, which causes partly reversible inhibition of cholinesterase [*2*]. The spontaneous return of function within an hour after sarin administration suggests that the inhibition of muscle cholinesterase by sarin may be to some extent spontaneously reversible during this time. Part of the muscle cholinesterase remains inhibited, however, as indicated by the increased effect of a second dose of sarin administered following complete recovery of neuromuscular transmission. (Fig. 5.)

Mechanism of Neuromuscular Block Produced by Large Doses of 2-PAM. The concentration of 2-PAM necessary to produce neuromuscular block in man was approximately 2×10^{-3} M. This is also the concentration of 2-PAM that inhibited cholinesterase, including that of muscle, *in vitro*. DAM, which had no appreciable anticholinesterase activity *in vitro*, had no neuromuscular blocking action. These observations suggest that the neuromuscular blocking action of 2-PAM may be due to its anticholinesterase activity. The occurrence of progressive depression of successive potentials evoked by repetitive nerve stimuli during the block is compatible with this mechanism, although such decrement also occurs in some other types of neuromuscular block. The neuromuscular blocking action of 2-PAM was enhanced following the administration of anticholinesterase compound, even after complete restoration of neuromuscular transmission. This suggests that PAM may produce neuromuscular block by virtue of either anticholinesterase or direct depolarizing action. The latter seems less likely, as the blocking action of 2-PAM was not enhanced by the prior administration of succinylcholine, which does produce a depolarizing type of block. Furthermore, 2-PAM has been found in experimental animals to have no effect on neuromuscular block due to d-tubocurarine [*22*]. While the transient nature of the neuromuscular block produced by 2-PAM made it difficult to study in detail, certain observations did not support the concept that it is due to cholinesterase inactivation. The prompt depressant action of acetylcholine [*31*] was unchanged during the block, and the block was not intensified by neostigmine. Furthermore, 2-PAM has been found to produce neuromuscular block in experimental animals in concentrations at which its anticholinesterase activity is minimal [*22*], as well as changes in the end-plate potential which are not suggestive of such activity [*32*]. The cause of the neuromuscular block produced by 2-PAM is, therefore, not clear. Holmes and Robins [*22*] have suggested that this compound may have a direct toxic action on the muscle fiber, since the response of muscle to direct stimulation was reduced in the experimental animal, and conduction velocity slowed. In man, large doses of 2-PAM produced slightly more depression of the tension response of muscle to nerve stimulation than of the action potential response, but the difference was not marked. (Fig. 1.)

The dose of 2-PAM that will produce systemic weakness in man is not yet known. Since the difference between the intra-arterial dose that reversed anticholinesterase neuromuscular block and that which produced local weakness was 2,000 fold, there is probably a wide range between the systemic therapeutic dose and that which would produce weakness. However, the neuromuscular blocking action of 2-PAM is enhanced by the prior administration of anticholinesterase compound so that the possibility of production of neuromuscular block by 2-PAM during the management of anticholinesterase poisoning cannot be ignored.

Anticholinesterase neuromuscular block can be reversed by competitive blocking agents such as d-tubocurarine which inhibit the action of acetylcholine on the motor end-plates but, in contrast to the oximes, the difference between the effective dose and that which produces neuromuscular block is so small (approximately 1 to 25 [*11*]) that use of these agents in the management of anticholinesterase intoxication is too hazardous [*3*].

Management of Anticholinesterase Intoxication. The management of intoxication due to anticholinesterase compounds has hitherto relied upon administration of large doses of atropine to diminish muscarine-like and central neural manifestations, and upon aspiration of bronchial and salivary secretions, maintenance of an adequate airway, and artificial respiration when needed [*3*]. In patients with impaired breathing, atropine may facilitate respiratory exchange to some extent by suppressing bronchial and

salivary secretion and bronchoconstriction and probably by diminishing central depression of respiration, but it does not appreciably restore strength to the respiratory or pharyngeal muscles [*3*]. In contrast, the intravenous administration of adequate doses of 2-PAM or DAM does improve muscular strength, and prompt administration of adequate doses should diminish the necessity for, or duration of, mechanical measures to sustain respiration. However, since weakness may develop rapidly in the course of severe anticholinesterase intoxication, these measures will undoubtedly still be needed in many patients. The intravenous administration of adequate doses of atropine also remains an important adjunct in treatment, since the oximes do not appear to reverse the muscarine-like manifestations of anticholinesterase intoxication, and their ability to reverse central neural effects remains to be demonstrated. Koelle has shown that 2-PAM can reverse the inhibition of ganglion cholinesterase by DFP, and it is possible that reversal of central neural effects of anticholinesterase compounds may occur and that the requirement for atropine may be diminished. In experimental animals the therapeutic effects of 2-PAM and atropine in sarin intoxication appear to be more than additive [*23*].

The studies that were carried out did not clearly demonstrate the optimal dose of oxime, or the superiority of either of the oximes studied. DAM produced more local and systemic symptoms and reduction in blood pressure than 2-PAM, and had to be injected slowly. This may prove to be a disadvantage, since rapidity of injection of either compound appeared to be advantageous in reversing anticholinesterase neuromuscular block. It is not clear whether the greater efficacy of oxime following intra-arterial than intravenous administration is due to more rapid or to more marked increase in concentration at the neuromuscular junction. The intra-arterial injection of 0.05 mg. of oxime probably resulted in a peak concentration of approximately 1 mg./L. in the injected hand, while the intravenous injection of 2,000 mg., if distributed throughout an extracellular volume of 15 L., would yield a concentration of 130 mg./L. Preliminary studies indicate that the plasma concentration of oxime determined colorimetrically following intravenous injection is about one-third of this value, but it has not yet been ascertained if this reflects the concentration of active compound. In experimental animals DAM has been found to be rapidly removed from the blood stream, and to be metabolized by the liver [*25*]. The clinical observations suggest that repeated intravenous administration of oxime may be necessary in the management of severe anticholinesterase intoxication, that intramuscular and oral routes of administration will probably not be effective, and that 2-PAM may prove more effective than DAM because it can be injected more rapidly. 2-PAM has the disadvantage that very high concentrations produce neuromuscular block, which is enhanced by prior exposure to anticholinesterase compound. The dose of this compound that will produce generalized weakness is not known, but is undoubtedly many times the therapeutic dose. No advantage was found in the simultaneous or successive administration of the two oximes.

It is not clear whether or not oxime must be administered shortly after absorption of the anticholinesterase compound in order to be maximally effective. Reversal of neuromuscular block occurred when oxime was administered four hours after absorption of anticholinesterase compound, and reversal of plasma and red blood cell cholinesterase occurred when oxime was administered twenty-four hours after absorption of an organophosphorus anticholinesterase agent. Rapid treatment is desirable to prevent progression of intoxication, but the time limit for maximal efficacy remains to be determined.

The toxic doses and nature of the toxic effects of 2-PAM and DAM in man are not known. The LD_{50} of 2-PAM has been found to be 190 mg. per Kg. in mice, and of DAM 900 mg. per Kg. [*25*]. The nature of the acute toxicity of these compounds is still obscure even in experimental animals, but in mice central nervous system depression appears to be the immediate cause of death. Toxic symptoms were encountered by these authors following the continued administration of 390 mg. of DAM per Kg. per day. No consistent pathologic changes were demonstrated.

While the optimal and toxic doses of oxime, and the optimal frequency of administration, are not yet known, the doses employed in this study were well tolerated and produced moderate relief from weakness due to organophosphorus or quaternary ammonium anticholinesterase compounds. The following procedure is therefore recommended for the management of intoxication due to these compounds:

1. *Termination of exposure:* removal of casualty or use of gas mask if atmosphere is contaminated; removal of contaminated clothing; washing of contaminated skin or eyes with copious amounts of water; gastric lavage if ingestion has occurred; application of tourniquet if exposure is percutaneous or intramuscular [*3*].

2. *Removal of secretions and maintenance of patent airway:* prone position with head down and to one side, mandible elevated, and tongue pulled forward; clearing of mouth and pharynx with finger or suction; oropharyngeal or nasopharyngeal airway in unconscious or flaccid patients, or endotracheal intubation if airway obstruction persists [*3,34*].

3. *Artificial respiration when necessary:* mouth-to-mouth, mask-to-mask, bellows, or mechanical [*34*].

4. *Atropine administration:* in severe intoxication, particularly by organophosphorus compounds, 2 to 4 mg. intravenously, followed by 2 mg. every three to eight minutes until muscarine-like symptoms disappear, and whenever they reappear; a total of 24 to 48 mg. may be required the first day; in less severe intoxication 2 mg. intravenously or intramuscularly, repeated at twenty-minute intervals until muscarine-like symptoms are relieved; maintenance of a mild degree of atropinization for twenty-four to forty-eight hours [*3,35*].

5. *Oxime administration:* in severe intoxication 2,000 mg. of 2-PAM (500 mg./minute) or DAM (200 mg./minute) intravenously; ? repetition of dose if weakness is not relieved, or recurs; in moderate intoxication 1,000 mg. intravenously, repeated if weakness is not relieved or recurs.

6. *Alleviation of convulsions if these interfere with respiration:* trimethadione (tridione®), 1 gm. intravenously every fifteen minutes up to a maximum of 5 gm., or sodium thiopental (2.5 per cent solution) intravenously [*3*].

SUMMARY

The oximes, pyridine-2-aldoxime (2-PAM) and diacetyl monoxime (DAM), protected against the inhibition of human cholinesterase enzymes by organophosphorus and quaternary ammonium anticholinesterase compounds *in vitro*, and 2-PAM reversed this inhibition.

The oximes reversed neuromuscular block and plasma and red blood cell cholinesterase inhibition produced in normal subjects by the administration of sarin, neostigmine, bis-neostigmine, pyridostigmine, bis-pyridostigmine and ambenomium. The intravenous dose required to alleviate generalized weakness was 1,000 to 2,000 mg. These doses did not relieve the muscarine-like effects of the anticholinesterase compounds, and their influence on central neural effects was not pronounced. DAM produced local burning and mild systemic symptoms. 2-PAM produced a transient, local neuromuscular block following the intra-arterial injection of high concentrations. This was enhanced by the prior injection of anticholinesterase compound.

2-PAM and DAM are valuable adjuncts to atropine in the management of anticholinesterase intoxication, and should diminish the necessity for, or duration of, artificial respiration and endotracheal intubation.

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REFERENCES

1. Grob, D., Garlick, W. L. and Harvey, A. M. The toxic effects in man of the anticholinesterase insecticide parathion (p-nitrophenyl diethyl thionophosphate). *Bull. Johns Hopkins Hosp.*, 87: 106, 1950.
2. Grob, D. and Harvey, A. M. Observations on the effects of tetraethyl pyrophosphate (TEPP) in man and on its use in the treatment of myasthenia gravis. *Bull. Johns Hopkins Hosp.*, 84: 543, 1949.
3. Grob, D. The manifestations and treatment of poisoning due to nerve gas and other organic phosphorus anticholinesterase compounds. *Arch. Int. Med.*, 98: 221, 1956.
4. Grob, D. and Harvey, J. C. Effects in man of the anticholinesterase compound sarin. (isopropyl methyl phosphonofluoridate). (In press.)
5. Rowland, L. P., Korengold, M. C., Jaffe, I. A., Berg, L. and Shy, G. M. Prostigmin-induced muscle weakness in myasthenia gravis patients. *Neurology*, 5: 89, 1955.
6. Pateisky, K., Kraupp, O. and Stumpf, C. Klinische Erfahrungen mit Hexamethylen-bis-(N-methyl-carbaminoyl-m-trimethylammoniumphenol) (BC 40) bei Myasthenia gravis pseudoparalytica. *Wien. klin. Wchnschr.*, 67: 578, 1955.
7. Osserman, K. E., Teng, P. and Kaplan, L. I. Studies in myasthenia gravis. Preliminary report on therapy with Mestinon bromide. *J. A. M. A.*, 155: 961, 1954.
8. Schwab, R. S., Marshall, C. K. and Timberlake, W. Win 8077 in the treatment of myasthenia gravis. *J. A. M. A.*, 158: 625, 1955.
9. Grob, D., Lilienthal, J. L., Jr., Harvey, A. M., Jones, B. F., Langworthy, O. R. and Talbot, S. A. The administration of di-isopropyl fluorophosphate (DFP) to man. I–IV. Effect on cholinesterase; systemic effects; effect on intestinal

motility, central nervous system, and myasthenia gravis. *Bull. Johns Hopkins Hosp.*, 81: 217, 1947.

10. Schulman, S., Rider, J. A. and Richter, R. B. Use of octamethyl pyrophosphoramide in the treatment of myasthenia gravis. *J. A. M. A.*, 152: 1,707, 1953.
11. Grob, D., Johns, R. J. and Harvey, A. M. Studies in neuromuscular function. v. Effects of anticholinesterase compounds, d-tubocurarine, and decamethonium in normal subjects. *Bull. Johns Hopkins Hosp.*, 99: 195, 1956.
12. Wilson, I. B. and Meislich, E. K. Reactivation of acetylcholinesterase inhibited by alkylphosphates. *J. Am. Chem. Soc.*, 75: 4,628, 1953.
13. Wilson, I. B. Reactivation of human serum esterase inhibited by alkylphosphates. *J. Am. Chem. Soc.*, 77: 2,383, 1955.
14. Wilson, I. B. and Ginsburg, S. Reactivation of acetylcholinesterase inhibited by alkylphosphates. *Arch. Biochem.*, 54: 569, 1955.
15. Wilson, I. B. and Ginsburg, S. A powerful reactivator of alkylphosphate-inhibited acetylcholinesterase. *Biochem. et Biophys. Acta*, 18: 168, 1955.
16. Hackley, B. E., Jr., Plapinger, R., Stolberg, M. and Wagner-Jauregg, T. Acceleration of the hydrolysis of organic fluorophosphates and fluorophosphonates with hydroxamic acids. *J. Am. Chem. Soc.*, 77: 3,651, 1955.
17. Jandorf, B. J., Crowell, E. A. and Levin, A. P. Role of hydroxamic acids in prevention and reversal of cholinesterase inactivation by DEP and sarin. *Federation Proc.*, 14: 231, 1955.
18. Childs, A. F., Davies, D. R., Green, A. L. and Rutland, J. P. The reactivation by oximes and hydroxamic acids of cholinesterase inhibited by organo-phosphorus compounds. *Brit. J. Pharmacol.*, 10: 462, 1955.
19. Kewitz, H. A specific antidote against lethal alkyl phosphate intoxication. iii. Repair of chemical lesion. *Arch. Biochem.*, 66: 263, 1957.
20. Kewitz, H. and Nachmansohn, D. A specific antidote against lethal alkyl phosphate intoxication. iv. Effects in brain. *Arch. Biochem.*, 66: 271, 1957.
21. Roy, B. B. and Kuperman, A. S. Reversal of the actions of tetraethyl pyrophosphate in surviving mammalian tissue. *Proc. Soc. Exper. Biol. & Med.*, 89: 255, 1955.
22. Holmes, R. and Robins, E. L. The reversal by oximes of neuromuscular block produced by anticholinesterases. *Brit. J. Pharmacol.*, 10: 490, 1955.
23. Brown, R. V., Kunkel, A. M., Somers, L. M. and Wills, J. H. Pyridine-2-aldoxime methiodide in the treatment of poisoning by sarin or tabun, with notes on its pharmacology. *J. Pharmacol. & Exper. Therap.* (In press.)
24. Wills, J. H., Kunkel, A. M., Brown, R. V., and Groblewski, G. E. Pyridine-2-aldoxime methiodide and poisoning by anticholinesterases. *Science*, 125: 743, 1957.
25. Dultz, L., Epstein, M. A., Freeman, G., Gray, E. H. and Weil, W. B. Studies on a group of oximes as therapeutic compounds in sarin poisonings. *J. Pharmacol. & Exper. Therap.*, 119: 522, 1957.
26. Kewitz, H., Wilson, I. B. and Nachmansohn, D. A specific antidote against lethal alkyl phosphate intoxication. ii. Antidotal properties. *Arch. Biochem.*, 64: 456, 1956.
27. Michel, H. O. An electrometric method for the determination of red blood cell and plasma cholinesterase activity. *J. Lab. & Clin. Med.*, 34: 1,564, 1949.
28. Grob, D., Johns, R. J. and Harvey, A. M. Studies in neuromuscular function. i. Introduction and methods. *Bull. Johns Hopkins Hosp.*, 99: 115, 1956.
29. Johns, R. J. A transducer tube myograph for use in man. *Electroencephalog. & Clin. Neurophysiol.*, 8: 339, 1956.
30. Wilson, I. B. Mechanism of Enzyme Action, p. 642. Edited by McElroy and Glass. Baltimore, 1954. Johns Hopkins Press.
31. Grob, D., Johns, R. J. and Harvey, A. M. Studies in neuromuscular function. iii. Stimulating and depressant effects of acetylcholine and choline in normal subjects. *Bull. Johns Hopkins Hosp.*, 99: 136, 1956.
32. Wagley, P. F. A study of end-plate potentials, i. Observations on the actions of certain oximes. *Bull. Johns Hopkins Hosp.* (In press.)
33. Koelle, G. B. Histochemical demonstration of reactivation of acetylcholinesterase *in vivo*. *Science*, 125: 1,195, 1957.
34. Elam, J. O., Clements, J. A., Brown, E. S. and Elton, N. W. Artificial respiration for nerve gas casualty. *U. S. Armed Forces M. J.*, 7: 797, 1956.
35. Grob, D. Manifestations and treatment of nerve gas poisoning in man. *U. S. Armed Forces M. J.*, 7: 781, 1956.

Use of Oximes in the Treatment of Intoxication by Anticholinesterase Compounds in Patients with Myasthenia Gravis*

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INTOXICATION by anticholinesterase compounds frequently occurs in patients with myasthenia gravis following the administration of excessive doses. In the management of this disease graded doses of an anticholinesterase compound are administered until a maximal level of strength is attained in the affected muscles [*1*]. Unfortunately, in patients with severe myasthenia the maximal strength attained may be far below normal. Increasing doses may result in no further increase in strength, and excessive drug may produce generalized weakness [*2*]. This has been termed "cholinergic crisis" by some, to distinguish it from weakness due to the disease itself ("myasthenic crisis"). If the cholinergic crisis is severe, death may occur as a result of paralysis of the muscles of respiration and of the pharynx and tongue unless artificial respiration and an open airway are maintained until spontaneous recovery occurs. There has been no clinically useful means of accelerating recovery from this neuromuscular block.

The weakness that occurs in myasthenic patients following excessive doses of anticholinesterase compounds is similar to that which occurs in normal subjects. However, whereas generalized muscular fasciculations and muscarine-like symptoms (nausea, vomiting, abdominal cramps, diarrhea, sweating) invariably develop in normal subjects, fasciculations are frequently absent or minimal in patients with severe myasthenia, and muscarine-like symptoms may be either pronounced, mild or absent. Furthermore, the prior administration of atropine may suppress the muscarine-like symptoms. In the absence of these collateral signs of anticholinesterase intoxication it is sometimes difficult to determine whether an increase in weakness in a myasthenic patient is due to an overdose of anticholinesterase drug or to progression of the myasthenia and the need for more drug.

The longer the duration of action of an anticholinesterase compound, the more prolonged and even is the increase in strength which it produces in myasthenic patients, and the longer may be the interval between doses, but the likelihood of cumulation of repeated doses and of administration of an overdose are also greater. Thus the long-acting organophosphorus anticholinesterase compounds such as tetraethylpyrophosphate (TEPP) [*3*] and octamethyl pyrophosphoramide (OMPA) [*4*], and long-acting quaternary ammonium compounds such as bis-neostigmine (BC-40) [*5*] and bis-pyridostigmine or hexamarium (BC-51) are most likely to produce cumulation and overdose. Short-acting quaternary ammonium compounds such as neostigmine are least likely to have this effect, unless administered in conjunction with one of the longer acting drugs. While neostigmine is safest in this regard, its short action results in less even strength and requires more frequent administration. If excessive doses are given, even neostigmine may produce weakness [*6*]. Quaternary ammonium compounds with an intermediate duration of action, such as pyridostigmine [*7*] and ambenomium [*8*], usually produce more satisfactory regulation because they afford a

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† Kenny Foundation Scholar.

more sustained increase in strength than does neostigmine, but they are also intermediate with regard to danger of overdose.

In the preceding communication [*9*] evidence was provided that cholinesterase inhibition and neuromuscular block produced by quaternary ammonium or organophosphorus anticholinesterase compounds in normal subjects are reversed following the administration of pyridine-2-aldoxime methiodide (2-PAM) or diacetyl monoxime (DAM). This communication will present evidence that these oximes reverse the action of anticholinesterase compounds in myasthenic patients as well, and are of clinical value in the management of weakness due to overtreatment with these compounds.

PROCEDURE

The procedure was described in the preceding communication [*9*]. The patients studied had moderate to severe generalized myasthenia gravis, requiring from 180 to 540 mg. of pyridostigmine every three to four hours when awake. Studies were initiated when the patients were in the basal state, having received no medication for eight to twelve hours. The anticholinesterase compounds that were administered included the quaternary ammonium compounds neostigmine (prostigmin®),* bis-neostigmine (BC-40),† pyridostigmine (mestinon®),* bis-pyridostigmine (BC-51),† and ambenomium (mytelase®),‡ and the organophosphorus compounds sarin (isopropyl methyl phosphonofluoridate) and octamethyl pyrophosphoramide (OMPA).§ The effect of oxime on alterations in neuromuscular function produced by sarin is presented in detail. (Figs. 1 and 2.) The effect on alterations produced by the other anticholinesterase compounds was similar.

RESULTS

Effects of Administration of Oximes. The systemic and local effects of the oximes following intra-arterial and intravenous administration were the same as in normal subjects, including their effect on neuromuscular transmission and their lack of effect on the prompt depressant action of acetylcholine.

Effect of Administration of Oximes on the Actions of Anticholinesterase Compounds. Reversal by intra-arterial 2-PAM and DAM of alterations in neuromuscular transmission produced by anticholinesterase compounds: The intra-arterial injection of 0.05 to 0.2 mg. of either oxime resulted in striking reversal of alterations in neuromuscular transmission produced by neostigmine, bis-neostigmine, pyridostigmine, bis-pyridostigmine, ambenomium and sarin. Prior to the administration of the anticholinesterase agent, the patients manifested the defect in neuromuscular transmission characteristic of myasthenia gravis [*10*]; there was a progressive decline (decrement) in the amplitude of successive muscle action potentials evoked by nerve stimuli delivered at 40 msec. intervals. (Fig. 1.) Following the intra-arterial administration of sufficient anticholinesterase agent to repair this defect in transmission, as indicated by an increase in the amplitude of the evoked potentials and tension, the injection of 2-PAM or DAM resulted in depression of the potentials and tension, which were restored to their initial amplitude. (Fig. 1.) The subsequent injection of 2-PAM resulted in further depression of neuromuscular transmission. As in normal subjects, the depressant effect of this drug was enhanced by the prior administration of an anticholinesterase compound.

When an excess of anticholinesterase compound was administered, repair of the myasthenic defect in neuromuscular transmission was followed by progressive depression of the evoked potentials, accompanied in some instances by repetitive firing of the initial potential of a train. (Fig. 2.) This depression was similar to the effect of anticholinesterase compounds in normal subjects [*11*], except that larger doses were required. The administration of 0.05 mg. of 2-PAM or DAM resulted in prompt reversal of this block, as in normal subjects, with increase in amplitude of the evoked potentials and tension. (Fig. 2.) After these had been restored to their maximal level the further administration of oxime resulted in depression of transmission, which was restored to or below the level that had existed prior to administration of any anticholinesterase compound.

As in normal subjects, the effects of 2-PAM and DAM were similar, except for the more marked depressant effect of large doses of 2-PAM on neuromuscular transmission when administered alone or, more particularly, following an anticholinesterase compound. As in normal

* Supplied by Roche Laboratories, Nutley, New Jersey.

† Supplied by Merck Sharp & Dohme, Philadelphia, Pennsylvania.

‡ Supplied by Winthrop Labs., New York City.

§ Supplied by Eli Lilly & Co., Indianapolis, Indiana.

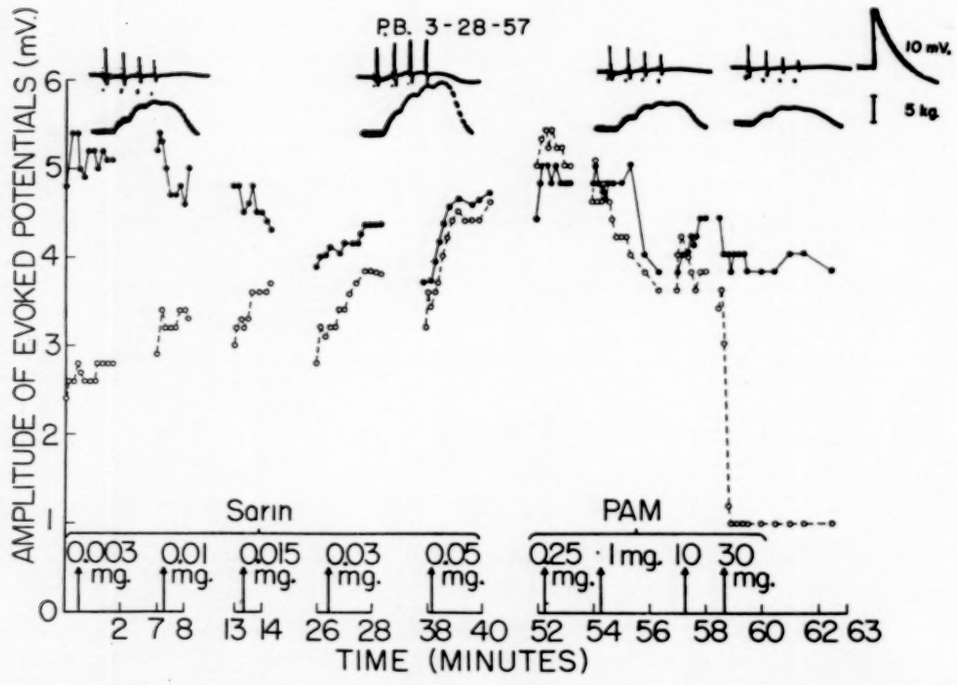

FIG. 1. Reversal by 2-PAM of the reparative effect of sarin on evoked muscle action potentials and tension. The amplitude of the first (●———●) and fourth (○———○) potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted, and illustrative potentials (mV.) and tension (kg.) recorded above. Injections were intra-arterial.

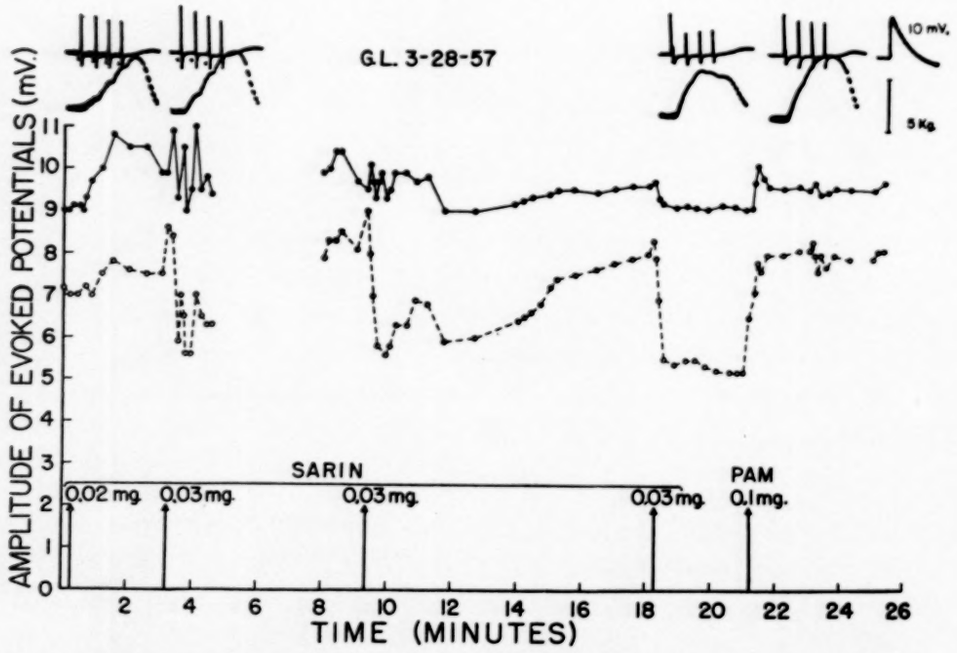

FIG. 2. Reversal by 2-PAM of the depressant effect of excessive sarin on evoked muscle action potentials and tension. The latter had been preceded by the reparative effect of prior injections of sarin. The amplitude of the first (●———●) and fourth (○———○) potentials in response to a train of four nerve stimuli (40 msec. apart) evoked every five seconds has been plotted, and illustrative potentials (mV.) and tension (kg.) recorded above. Injections were intra-arterial.

subjects, the effect of the simultaneous or successive injection of 2-PAM and DAM was additive.

Reversal by intravenous 2-PAM and DAM of changes in general strength produced by anticholinesterase compounds: The effect of the oximes was

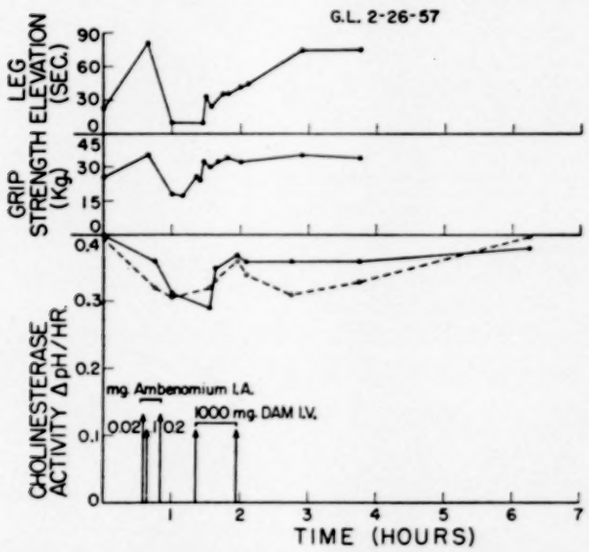

FIG. 3. Reversal by DAM of generalized weakness and plasma (●——●) and red blood cell (○——○) cholinesterase inhibition produced by excess ambenomium. The weakness was preceded by an increase in strength resulting from the prior injection of smaller doses of ambenomium.

similar to the local effect of smaller doses administered intra-arterially. The intravenous injection of sufficient oxime resulted in reversal of the change in general strength produced by each of the anticholinesterase compounds studied, regardless of whether this had been reparative or depressant. Prior to the administration of any drug, the patients had moderate to severe generalized weakness. When sufficient anticholinesterase compound was administered to produce an increase in strength, the intravenous injection of 300 to 2,000 mg. of 2-PAM or DAM resulted in a decrease in strength, with return of the characteristic myasthenic symptoms. (Table 1.) These symptoms were severe only in patients who had a severe degree of weakness in the basal state. When an excess of anticholinesterase compound had been administered, resulting in generalized weakness accompanied in most patients by muscular fasciculations, the administration of this dose of oxime produced moderate improvement in strength, which gradually attained the maximal level that had been present following optimal doses of the anticholinesterase agent. (Fig. 3 and Table 1.) The degree of change and the time course were similar to those in normal subjects. As in normal subjects, there was a less striking diminution in muscular fasciculations and no change in muscarine-like

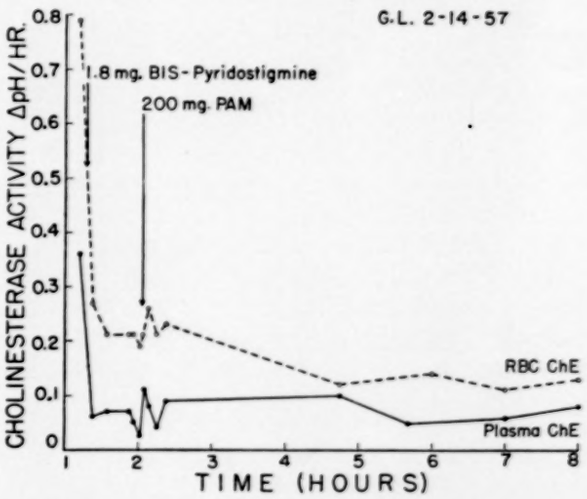

FIG. 4. Transient and slight reversal by 2-PAM (intravenous) of plasma and red blood cell cholinesterase inhibition produced by bis-pyridostigmine (intra-arterial).

symptoms. The further administration of oxime, after recovery of strength, resulted in some instances in a decrease in strength, with return of the characteristic myasthenic symptoms.

Reversal by 2-PAM and DAM of plasma and red blood cell cholinesterase inhibition produced by anticholinesterase compounds: This was the same as in normal subjects. (Figs. 3 and 4 and Table 1.)

CASE REPORT

G. L., a seventy-six year old man with moderately severe generalized myasthenia gravis, received 0.1 mg. of ambenomium intra-arterially when in the basal state. There was an increase in general strength to the maximal level attainable following optimal doses of anticholinesterase medication. (Fig. 3.) The patient then received an additional 0.2 mg. of ambenomium intra-arterially. Ten minutes later he began to have difficulty swallowing, dysarthria developed, and a marked decrease in general strength was noted. He was barely able to elevate his head off the pillow or raise either leg. There was slight respiratory distress, and moderate muscular fasciculations which were most pronounced in the legs. Twenty minutes after the injection of ambenomium the patient was given 1,000 mg. of DAM intravenously over a period of thirty minutes in increments of 100 mg. injected at the rate of 10 mg. per second. Following each injection there was a burning sensation in the injected

TABLE I

EFFECT OF ADMINISTRATION OF 2-PAM AND DAM ON THE ALTERATIONS IN GENERAL STRENGTH AND CHOLINESTERASE ACTIVITY (PLASMA AND RED BLOOD CELL) PRODUCED BY ANTICHOLINESTERASE COMPOUNDS

Patient	Anti-cholinesterase Compound	Dose (mg.)	Route	Effect of Anticholinesterase Compound			Oxime i.v.* (mg.)	Effect of Oxime On:		Cholinesterase Activity (ΔpH/hr.)					
										Plasma			Red Blood Cell		
				Gastrointestinal Symptoms	Strength	Fasciculation (Increase)		Strength	Fasciculation (Decrease)	Control	Before Oxime	Increase After Oxime	Control	Before Oxime	Increase After Oxime
PAM															
H. F.	Neostigmine	180	p.o.	++++	↓++++	+++	1000	↑++	+	0.72	0.17	0.01	0.65	0.17	0
D. K.	Neostigmine	5.5	i.a.	+	↑++	0	60	0	0	0.35	0.18	0.09	0.42	0.39	0.03
G. L.	Neostigmine	3.5	i.a.	+	↑++	+	150	0	±	....	....	....	....	....	....
G. L.	Bis-pyridostigmine	0.8	i.a.	0	↑++	+	160	0	±	0.36	0.03	0.08	0.79	0.19	0.05
G. L.	Bis-pyridostigmine	1.8	i.a.	+	↑++	+	650	0	+	0.40	0	0	0.74	0.14	0.02
E. K.	Bis-pyridostigmine	2	i.a.	0	↑++	0	60	0	..	0.42	0.08	0.06	0.46	0.22	0.07
M. O.	OMPA	33	p.o.	0	↑++	0	1000	↓++	..	0.82	0.44	0.03	0.63	0.32	0.04
G. L.	Sarin	0.18	i.a.	0	↑+	0	550	↓+	..	0.40	0.33	0.07	0.78	0.62	0.16
P. B.	Sarin	0.18	i.a.	0	↑+	0	550	↓+	..	0.89	0.52	0.12	0.68	0.42	0.14
DAM															
E. K.	Neostigmine	5.5	i.a.	+	↑++	0	60	0	0	0.35	0.18	0.15	0.42	0.39	0.03
G. L.	Neostigmine	2.6	i.a.	+	↑++	++	100	0	±	....	....	....	....	....	....
G. L.	Bis-pyridostigmine	1.8	i.a.	+	↑++	++	65	0	0	0.40	0	0	0.74	0.13	0.02
D. K.	Bis-pyridostigmine	2	i.a.	0	↑++	0	60	0	..	0.42	0.08	0.03	0.46	0.16	0.03
G. L.	Ambenomium	0.3	i.a.	+	↓++	+++	1000	↑++	+	0.40	0.29	0.08	0.74	0.32	0.04
G. L.	Ambenomium	0.4	i.a.	+	↓++	+++	500	↑+	0	....	....	....	....	....	....
M. O.	OMPA	33	p.o.	0	↑+	0	700	0	..	0.82	0.32	0.02	0.63	0.40	0.02
M. O.	OMPA	17	p.o.	0	↑+	0	1000	↓+	..	....	0.26	0.02	....	0.29	0.01
G. L.	Sarin	0.18	i.a.	0	↑+	0	220	0	..	0.40	0.28	0.04	0.78	0.51	0.11
P. B.	Sarin	0.18	i.a.	0	↑+	0	275	0	..	0.89	0.75	0.03	0.68	0.52	0.05

NOTE: The symbol ↑ denotes an increase, and ↓ a decrease. The number of + signs denotes degree of change (range + to ++++).
* Intravenously.

antecubital vein, radiating to the axilla, but there were no systemic symptoms or change in blood pressure. Following the injection of 300 mg. of DAM there was slight improvement in swallowing, speech and respiration. After 500 mg. there was moderate improvement in general strength. After 1,000 mg. his strength returned to the level that had existed following the initial injection of 0.1 mg. ambenomium and remained at this level for five hours. Fasciculations were only slightly diminished. There was transient restoration of plasma and red blood cell cholinesterase activity, which had been moderately depressed following the administration of ambenomium.

COMMENTS

The defect in neuromuscular transmission in myasthenia gravis appears to be due to a competitive type of block which inhibits the motor end-plate depolarizing action of acetylcholine released from the motor nerve endings [*12*]. Following the administration of an anticholinesterase compound, partial or complete repair of this defect occurs, presumably due to the inhibition of muscle cholinesterase and the resultant accumulation of sufficient acetylcholine at the motor end-plates to overcome the competitive block. The further administration of an excess of anticholinesterase compound results, in most patients, in depression of neuromuscular transmission, probably because sufficient acetylcholine accumulates to cause persistent depolarization of the end-plate region [*12*]. The depression is similar to that observed in normal subjects except that more anticholinesterase compound usually is required, especially in patients with severe myasthenia. It is not clear

why depression may occur without prior restoration of neuromuscular transmission to normal.

The neuromuscular actions of anticholinesterase compounds were reversed by the oximes in patients with myasthenia gravis, as in normal subjects. The effect of this reversal depended on the prior action of the anticholinesterase compound, since neuromuscular function was restored toward its initial state. In normal subjects, neuromuscular function and strength were returned toward normal. In myasthenic patients, the effect depended on the status of the patient at the time of oxime administration. If sufficient anticholinesterase compound had been administered to depress function, this was restored to a more optimal level, but if function was optimal at the time of oxime administration, it was restored toward the basal level present prior to the administration of anticholinesterase compound; i.e., it was depressed.

While the oximes were equally effective in the treatment of anticholinesterase intoxication in myasthenic patients and normal subjects, more cautious administration was necessary in the former. Following the amelioration of weakness due to this intoxication by the administration of 1,000 mg. of 2-PAM or DAM, repetition of this dose had no effect on strength in normal subjects [*9*], but produced weakness in some patients with severe myasthenia. In these patients, overtreatment with oxime may convert a cholinergic crisis into a myasthenic crisis. It is therefore recommended that myasthenic patients suffering from anticholinesterase intoxication be titrated with successive 500 mg. doses of 2-PAM or DAM at five- to ten-minute intervals until strength is restored to the maximal level attained following the administration of optimal doses of anticholinesterase compound.

Systemic weakness produced by the oximes in myasthenic patients was due to reversal of the action of previously administered anticholinesterase compound. The local neuromuscular block produced by large doses of 2-PAM injected intra-arterially was no greater in these patients than in normal subjects. This block therefore is not attributable to competitive inhibition of the action of acetylcholine on the motor end-plate, since such inhibition is more marked in myasthenic patients [*13*].

Availability of the oximes should enable potent and long-acting anticholinesterase compounds to be employed in the management of myasthenia gravis with less danger from the effects of drug cumulation and overdose. The oximes may be used not only to reverse such effects but also to recognize them. In patients with severe myasthenia who are receiving large amounts of anticholinesterase medication it is sometimes difficult to determine whether an increase in weakness is due to overdose of medication or to the need for more drug. The effect of oxime administration is helpful in making this differentiation: an increase in strength indicates that the patient has become weak because of an excess of anticholinesterase drug, while a decrease indicates that the patient was either at an optimal level of strength or had not received enough anticholinesterase medication. The converse of this is seen following the intravenous injection of the short-acting anticholinesterase compound, edrophonium (tensilon®) * [*14*], when an increase in strength indicates insufficient anticholinesterase medication, and a decrease in strength an excess.

In the management of severe anticholinesterase intoxication in myasthenic patients, as in normal subjects, endotracheal intubation and artificial respiration may have to be performed before sufficient oxime can be administered to alleviate the weakness. However, it is likely that prompt injection of oxime will diminish the necessity for, and duration of, these measures. Administration of atropine (1 to 2 mg. intravenously or intramuscularly, repeated as necessary) will usually also be required, to relieve the muscarine-like manifestations of anticholinesterase intoxication, since these do not appear to be affected by the oximes.

SUMMARY

Administration of pyridine-2-aldoxime methiodide (2-PAM) or diacetyl monoxime (DAM) to patients with myasthenia gravis reversed the effects of the following anticholinesterase compounds on neuromuscular function, muscle strength, and plasma and red blood cell cholinesterase activity: neostigmine, bis-neostigmine, pyridostigmine, bis-pyridostigmine, ambenomium and sarin. The intravenous dose of these oximes that reversed the effect of anticholinesterase agents on general strength was 300 to 2,000 mg. Following the administration of sufficient anticholinesterase compound to repair the myasthenic defect in neuromuscular transmission and improve strength, the injection of oxime

* Supplied by Roche Laboratories, Nutley, New Jersey.

resulted in a decrease in transmission and strength, toward the basal level. Following an excess of anticholinesterase agent sufficient to cause neuromuscular block and weakness the injection of oxime resulted in improvement in neuromuscular function and strength. The subsequent injection of oxime resulted in some instances in a decrease in function and strength to the basal level.

The oximes have proved to be of value in the management in myasthenic patients of weakness due to overtreatment with anticholinesterase medication.

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REFERENCES

1. Grob, D. Myasthenia gravis. In: Current Therapy, p. 460. Edited by Conn, H. F. Philadelphia, 1956. W. B. Saunders Co.
2. Grob, D., Johns, R. J. and Harvey, A. M. Studies in neuromuscular function. VI. Effects of anticholinesterase compounds, d-tubocurarine, and decamethonium in patients with myasthenia gravis. *Bull. Johns Hopkins Hosp.*, 99: 219, 1956.
3. Grob, D. and Harvey, A. M. Observations on the effects of tetraethyl pyrophosphate (TEPP) in man and on its use in the treatment of myasthenia gravis. *Bull. Johns Hopkins Hosp.*, 84: 543, 1949.
4. Schulman, S., Rider, J. A. and Richter, R. B. Use of octamethyl pyrophosphoramide in the treatment of myasthenia gravis. *J. A. M. A.*, 152: 1707, 1953.
5. Pateisky, K., Kraupp, O. and Stumpf, C. Klinische Erfahrungen mit Hexamethylen-bis-(N-methylcarbaminoyl-m-trimethylammoniumphenol) (BC 40) bei Myasthenia gravis pseudoparalytica. *Wien. klin. Wchnschr.*, 67: 578, 1955.
6. Rowland, L. P., Korengold, M. C., Jaffe, I. A., Berg, L. and Shy, G. M. Prostigmin-induced muscle weakness in myasthenia gravis patients. *Neurology*, 5: 89, 1955.
7. Osserman, K. E., Teng, P. and Kaplan, L. I. Studies in myasthenia gravis. Preliminary report on therapy with mestinon bromide. *J. A. M. A.*, 155: 961, 1954.
8. Schwab, R. S., Marshall, C. K. and Timberlake, W. Win 8077 in the treatment of myasthenia gravis. *J. A. M. A.*, 158: 625, 1955.
9. Grob, D. and Johns, R. J. Use of oximes in the treatment of intoxication by anticholinesterase compounds in normal subjects. *Am. J. Med.*, 24: 497, 1958.
10. Johns, R. J., Grob, D. and Harvey, A. M. Studies in neuromuscular function. II. Effects of nerve stimulation in normal subjects and in patients with myasthenia gravis. *Bull. Johns Hopkins Hosp.*, 99: 125, 1956.
11. Grob, D., Johns, R. J. and Harvey, A. M. Studies in neuromuscular function. V. Effects of anticholinesterase compounds, d-tubocurarine, and decamethonium in normal subjects. *Bull. Johns Hopkins Hosp.*, 99: 195, 1956.
12. Grob, D., Johns, R. J. and Harvey, A. M. Studies in neuromuscular function. IV. Stimulating and depressant effects of acetylcholine and choline in patients with myasthenia gravis, and their relationship to the defect in neuromuscular transmission. *Bull. Johns Hopkins Hosp.*, 99: 153, 1956.
13. Grob, D., Johns, R. J. and Harvey, A. M. Studies in neuromuscular function. VI. Effects of anticholinesterase compounds, d-tubocurarine, and decamethonium in patients with myasthenia gravis. *Bull. Johns Hopkins Hosp.*, 99: 219, 1956.
14. Osserman, K. E. and Kaplan, L. I. Rapid diagnostic test for myasthenia gravis: Increased muscle strength, without fasciculations, after intravenous administration of edrophonium (tensilon) chloride. *J. A. M. A.*, 150: 265, 1952.

Bilateral Renal Cortical Necrosis*

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RENAL cortical necrosis is a pathological lesion associated with the clinical syndrome of abrupt onset of gross hematuria, oliguria, anuria and subsequent uremic syndrome. It is characterized by a bilateral, symmetrical, patchy-to-diffuse ischemic coagulative cortical necrosis with sparing of the renal medulla and an intact sub-capsular rim of cortical tissue. In the past it has been considered a rapidly and universally fatal disease. The present study concerns our experience with one patient who survived and two patients with fatal cortical necrosis who were among approximately 140 patients with acute oliguria studied in The Renal Laboratory over a four-year period. The clinical and experimental literature will be briefly discussed.

CASE REPORTS

CASE 1. A thirty-two year old Negro woman who had had four pregnancies and two abortions entered another hospital on June 10, 1954 with the diagnosis of abruptio placentae. On the day of admission she spontaneously delivered a five month non-viable infant and had severe uterine hemorrhage. The patient was in shock for an unknown length of time and received 2,000 ml. of whole blood. Albuminuria and hematuria were noted in addition to the anemia of blood loss. She had been known to have hypertension during a previous pregnancy which terminated with a spontaneous abortion at two months.

After recovery from shock, the patient was anuric and was transferred to Georgetown University Hospital on her fourth anuric day. Examination revealed an obese, comfortable, Negro woman. She was afebrile and had a pulse rate of 100/minute, respiratory rate of 16/minute and a blood pressure of 190/130 mm. Hg. The skin was warm, moist and well hydrated. The retinas showed moderate spasm of arterioles with venous compression, and were classified as grade 1. There was a grade 2 soft precordial systolic murmur.

The hematocrit was 28 per cent. There was a leukocytosis of 23,600/cu. mm. and a total eosinophil count of 6. The patient remained anuric for the succeeding ten days with a urine volume of 4 to 88 ml./twenty-four hours. The urine had a specific gravity of 1.010, a pH of 7.5, 4 plus albumin, many red blood cells, 1 to 5 white blood cells per high power field and many tubular epithelial cells. Examination of the blood on admission revealed the following: sodium, 120; potassium, 5; chloride, 80; carbon dioxide content, 11 mEq./L. The urea nitrogen was 125, the creatinine 18, the calcium 7.5 and the phosphorus 16 mg./100 ml. The serum alkaline phosphatase was 13.3 units. During ten days of anuria the patient showed only moderate clinical deterioration. She was treated with cation exchange resins, restriction of salt, protein, potassium and fluid.

The patient was dialyzed with a rotating drum artificial kidney on the eleventh day because of progressive uremia, and on the twentieth day because of hyperkalemia. The chemical results are summarized in Table 1. Four days following the first dialysis the daily urine volume increased successively to 127, 210, 310, 470, 350 and 290 ml. A uremic pericardial friction rub was noted before the second dialysis. A gram stain of the urine sediment revealed gram-negative rods, and a heavy culture of Aerobacter aerogenes was obtained. Albuminuria and pyuria persisted. On the twenty-second oliguric day pulmonary edema developed which responded to the usual measures. On the twenty-third oliguric day first-degree heart block was noted in the electrocardiogram. The patient was dialyzed for the third time on the twenty-fourth oliguric day (Table 1) and again showed marked clinical improvement following the procedure. Blood pressures were now ranging between 190/110 and 230/140 mm. Hg with recurring attacks of pulmonary edema. On the twenty-ninth oliguric day a grade 4 retinopathy with "sheen" was noted. The patient was then given a trial of ganglionic-blocking agents. On the thirtieth oliguric day the patient became totally anuric and comatose, with Cheyne-Stokes respiration. The blood pressure fell and the patient died on the thirty-fourth oliguric day. Postmortem examination revealed acute tracheobronchitis, bronchial pneumonia, acute pulmonary edema, left ventricular hypertrophy and bilateral cortical necrosis with calcification.

The right kidney weighed 160 gm., the left 180 gm. The cortical surface was smooth and mottled, with

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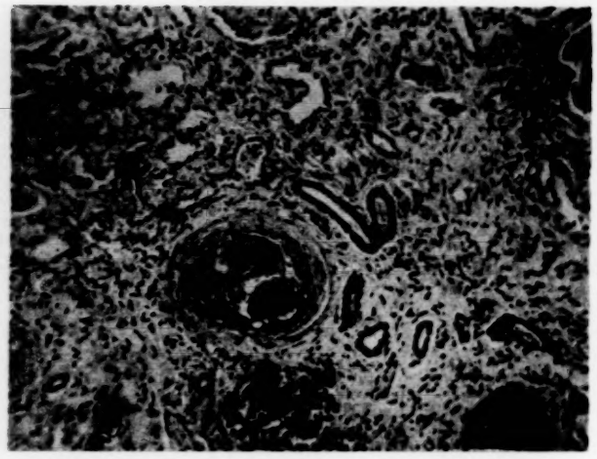

FIG. 1. Section through the renal cortical rim in Case 1 demonstrates hemorrhagic glomeruli, cellular infiltrate, proliferation of stroma and tubular degeneration. The cuff of periglomerular fibrosis is due to complicating pyelonephritis. Original magnification, × 200.

scattered small hemorrhagic and yellow areas. Gross examination of the cut section revealed the entire cortical rim to be yellow and necrotic except for a few scattered areas which were intensely congested. The cortical medullary line was distinct and the medulla showed intensely dark hemorrhagic streaks. Microscopic examination (Fig. 1) revealed a large number of blood vessels in the renal capsules, some containing blood. The normal architecture of the cortex was completely distorted except for a very thin sub-capsular area in which a few of the tubules were intact but filled with exudate containing a large number of leukocytes and homogeneous eosinophilic material. Glomeruli, tubules and interstitial tissue of the cortical area showed advanced necrosis with diffuse areas of calcification, hemorrhage and infiltrations with polymorphonuclear leukocytes and lymphocytes. There were a few intact glomeruli in scattered areas. These glomerular tufts were cellular but avascular. Glomeruli at the corticomedullary line preserved an outline of the tuft but represented only a pool of blood surrounded by a thin capsule. In the cortical area, the necrotic glomeruli were calcified. The tubules in the medulla were dilated and filled with eosinophilic casts and leukocytes. The lining epithelial cells were desquamated. In some areas there was calcification of the epithelial lining of the tubules. The blood vessels were dilated and engorged. Some showed marked thickening of the wall due to sclerosis and fibroblastic tissue proliferation. Some showed recent organized thrombosis with recanalization.

Comment: This patient fits into the so-called classic variety of cortical necrosis. She was a multiparous woman whose age was greater than

TABLE 1

CHEMICAL CHANGES BEFORE AND AFTER DIALYSIS

	Weight (kg.)	Blood Urea Nitrogen (mg./100 ml.)	Plasma				
			Creatinine (mg./100 ml.)	P (mg./100 ml.)	Na (mEq./L.)	K (mEq./L.)	Cl (mEq./L.)
Case I							
First dialysis:							
Pre	90	141	23.6	14.2	120	6.2	82
Post	90.4	50	8.7	5.1	135	4.0	98
Second dialysis:							
Pre	89	184	30.5	16.0	128	6.7	90
Post	.....	62	12.3	6.0	137	4.3	100
Third dialysis:							
Pre	.....	90	24.0	14.0	130	7.3	94
Post	.....	64	8.4	5.8	140	4.5	103
Case II							
First dialysis:							
Pre	93	190	22.2	6.0	130	7.3	99
Post	.....	73	12.0	3.0	140	4.0	104
Second dialysis:							
Pre	90.45	193	27.2	11.9	129	6.7	100
Post	.....	69	13.2	5.3	139	4.5	103

thirty years, and cortical necrosis occurred following shock due to abruptio placentae and blood loss. The thirty-four days of survival in such a patient whose course was complicated by pyelonephritis and bronchial pneumonia is, we believe, solely attributable to the availability of the artificial kidney. In viewing these kidneys, it is difficult to imagine how any function could take place in the presence of such massive destruction. Yet this patient was able to repair the few islands of incompletely damaged renal parenchyma and was able to achieve a urine volume as high as 480 ml./day. This would seem to permit a bare note of encouragement in the prolongation of life in such cases even when the diagnosis is known. It is notable that with perfusion through these damaged ischemic kidneys an acute and malignant form of hypertension developed, with cardiac hypertrophy and encephalopathy. This has also been a clinical feature of the early convalescent phase in other patients, in our experience, whose courses were not sufficiently well documented to warrant inclusion in this report. In a case seen with Dr. Arthur Merrill malignant hypertension also developed in the recovery phase.

Case II. A sixty-nine year old white man, with pre-existing hypertension, sustained a 12-inch scalp laceration in an auto accident on March 20, 1954. The patient was unconscious for five to ten minutes and bled profusely from the wound. He was admitted to another hospital where the laceration was sutured and whole blood replacement was given. On the second hospital day a urinary output of only 5 ml. was noted. On the fourth hospital day the non-protein nitrogen was 125, and the fasting blood sugar 102 mg./100 ml. The sodium was 134; the potassium, 6.2; the chloride, 102; and the carbon dioxide content, 16 mEq./L. There was a leukocytosis of 25,000/cu. mm., the hematocrit was 36 per cent. On the fifth hospital day a diffuse erythematous maculopapular rash was noted following blood transfusion. On the sixth hospital day the urinary output was 300 ml. with a specific gravity of 1.003 and 10 to 12 white blood cells and 12 to 15 red blood cells per high power field on microscopic examination. By the tenth hospital day the serum potassium had risen to 7.3 mEq./L. and the non-protein nitrogen to 190 mg./100 ml. while the hematocrit had fallen to 27. The patient was then transferred to Georgetown University Hospital as a candidate for dialysis. Physical examination at this time revealed a blood pressure of 165/90 mm. Hg; pulse, 100; respirations, 28; and rectal temperature, 100.6 F. degrees. The patient appeared acutely ill and was in a comatose state, exhibiting generalized body tremors with sporadic jerks of his extremities. Examination of the retinas showed arteriosclerosis and A-V compression.

Examination of the blood showed a hematocrit of 32 per cent, a sedimentation rate of 32 mm./hr., a leukocytosis of 33,200/cu. mm., and a total eosinophil count of 694. The urine was dark amber with a specific gravity of 1.008, a pH of 6, and 3 plus albuminuria. There were occasional white cells and 8 to 10 erythrocytes per high power field. A serologic test for syphilis was positive. The serum urea nitrogen was 190; the creatinine, 28; the calcium, 9.2; and the phosphorus, 6 mg./100 ml. The sodium was 130; the potassium, 7.3; the chloride, 99; and the carbon dioxide content, 9 mEq./L. The serum total bilirubin was 0.63 mg., the one-minute bilirubin 0.31 mg./100 ml. The platelet count was 158,000/cu. mm. The patient was dialyzed on the eleventh and twentieth anuric days, with good chemical (Table I) and clinical response. Anuria continued with daily urine outputs of 40 to 75 cc. On the twenty-second hospital day the patient was given digitalis for signs of congestive failure. Ventricular tachycardia, which had developed, persisted despite digitalization and the patient suddenly died on the twenty-fifth anuric day while an emergency electrocardiogram was being taken. Postmortem examination revealed bilateral renal cortical necrosis, nephrosclerosis, moderate coronary atherosclerosis, and chronic passive congestion of the liver and spleen. The kidneys were swollen and had mottled smooth external surfaces. The subcapsular portions of the cortex were slightly injected but intact. Immediately beneath these areas there was an irregular firm yellow, 3 to 4 ml. band of necrotic tissue involving the entire cortex and extending down into the columns of Bertin. Small petechial hemorrhages studded the border of this necrotic-like band. The cortical medullary line was distinct. There was a 2.5 cm. cyst extending from the left lateral surface in one kidney. Microscopic examination (Fig. 2) showed a thin rim of viable cortex beneath the capsule. Glomeruli in this area were normal or showed evidence of early hyalinization and fibrosis. The tubules were lined by atrophic epithelium and contained masses of leukocytes and blood in the lumens. The stroma was edematous and infiltrated by leukocytes and round cells. Beneath this zone there was an irregular band of necrotic hemorrhagic cortical tissue involving the entire cortex. (Fig. 3.) Except for the rim beneath the capsule and a small similar rim of cortex in the juxta-medullary region, all glomeruli, vessels, tubules and stroma in this band were necrotic and studded by minor fragments of nuclear debris. Many arcuate arteries contained organizing thrombi obliterating their lumens. Distal and collecting tubules contained blood and blood pigment casts. The medullary stroma was edematous and infiltrated by small round cells. There was moderate subintimal hyperplasia of the small arteries and arterioles.

FIG. 2. Low power view of the renal cortex in Case II. Note the intact cortex corticis, the zone of necrosis and the scattered areas of hemorrhage. Original magnification, × 12.

FIG. 3. Section through the necrotic renal cortex in Case II shows a ghostlike glomerulus and coagulated tubules. Original magnification, × 400.

Comment: This case exemplifies the unusual occurrence of cortical necrosis in an elderly man following what at the time appeared to be mild trauma. The patient suffered at the most five to ten minutes of unconsciousness following an automobile accident and the only visible injury was a superficial scalp laceration which produced considerable bleeding. Moreover, the patient's blood pressure was checked when he was admitted to the Emergency Room for repair of the laceration, and the injury was considered too minor to require hospitalization at the time of the accident. One blood transfusion was given without any recorded reaction. The subsequent course was one of continued severe anuria and progressive uremia. His maximum urine output in any one twenty-four-hour period was 75 ml. With the aid of hemodialysis he remained in remarkably good condition for twenty-five virtually anuric days and he was alert and comfortable up to the time he died with the sudden onset of ventricular tachycardia, apparently related to congestive failure and digitalization.

CASE III. A twenty-three year old white woman was admitted in coma. During the sixth month of her third pregnancy fever, nausea, vomiting, anorexia and diarrhea developed. Two weeks prior to admission she spontaneously delivered stillborn twins. Urinalysis then showed 2 plus proteinuria without abnormal sediment. She had a leukocytosis of 27,000. A lumbar puncture revealed an opening pressure of 380 mm. water. The non-protein nitrogen was 118 mg./100 ml.; the serum chloride, 108; carbon dioxide, 11; potassium, 4.3; and sodium, 156 mEq./L. The patient was oliguric up to three days prior to admission when diuresis began. The working diagnosis was necrotizing papillitis. The pyelonephritis increased in severity and the serum non-protein nitrogen increased to 268 mg./ml. Her condition worsened after the administration of 500 mg. of intravenous hydrocortisone and she was transferred for hemodialysis while deeply comatose. There was no past history of renal disease.

Physical examination revealed a well developed, white, obese woman in a comatose state. The respirations were 32; the blood pressure, 160/50 mm. Hg; and the pulse, 100. A diffuse generalized erythematous maculopapular rash was present. Hyperactive deep tendon reflexes with bilateral Babinski and Hoffmann signs were elicited. The oral cavity was lined with crusted blood. Laboratory work at the time of admission revealed a hemoglobin of 11 gm. per cent, hematocrit of 21 per cent and a white cell count of 37,700/cu. mm. with 10 bands. The icterus index was 7.5 and the total eosinophil count was 288. Urinalysis showed a specific gravity of 1.010, pH 6.5, 1 plus albumin with many clumps, white blood cells and an occasional red blood cell per high power field. Approximately 50 per cent of the leukocytes exhibited the glittering phenomenon, and occasional hyaline casts were present.

The patient was immediately dialyzed, with reversal of azotemia. There was marked clinical improvement following dialysis despite complications of acute exacerbations of Escherichia coli pyelonephritis, cardiac decompensation, atelectasis, thrombophlebitis and hypermetabolic syndrome [*1*]. On the thirty-fifth hospital day a percutaneous renal needle biopsy was performed with a modified Vim-Silverman needle. The patient was discharged on the

thirty-ninth hospital day and has remained well for approximately two years. No evidence of renal calcification has been found up to the present time.

The biopsy consisted of two pieces of tissue. One showed two glomeruli which were relatively normal in appearance except for slight hypercellularity. There was considerable atrophy of the tubules, many of which contained cellular colloid casts. There was chronic inflammatory cell infiltration of the stroma, and replacement of tubular parenchyma by inflammatory elements and scar tissue. The other piece consisted of a grossly distorted portion of renal parenchyma. There was almost total destruction of renal architecture. However, on careful examination there were ghost-like outlines of glomerular structure with a few remaining cells of Bowman's capsule. These glomeruli, of which there were six or eight possible examples on the slide, were separated by very pale staining non-cellular scar tissue resembling the end stage of coagulation necrosis. On the margin of this were some sclerosed blood vessels and a few tubules which showed regenerative changes.

Comment: This case represents the patchy type of cortical necrosis with a clinical course indistinguishable from that of acute renal insufficiency complicated by severe pyelonephritis. The patient recovered and the diagnosis was based entirely on the renal biopsy. It seems likely that this lesion may exist in other cases of acute tubular necrosis, particularly in the obstetrical group. It also seems likely that with increasing use of biopsy the diagnosis of patchy cortical necrosis will become more common.

REVIEW OF THE LITERATURE

Cortical necrosis was originally reported by Juhel-Renoy in 1886, in a young girl following an attack of scarlet fever [*2*]. Twelve years passed before another report appeared in the literature [*3*], thus establishing bilateral renal cortical necrosis as a new clinical entity. Since then many cases have been reported.

Ash in 1933 [*4*], in reviewing the literature, gathered together sixty-two cases, forty-four of which he considered to be verified. Most were associated with the complications of pregnancy and he noted a higher incidence in multiparous women over thirty-five years of age. The ages ranged between four years and sixty-nine years; seven of the cases reported were in children. He noted that the duration of the anuria had little to do with the final clinical outcome. He believed that the pathogenesis was an initial vasoconstriction of the renal vessels followed by a vasoparalysis and stasis with resultant necrosis of the distal cortical tissue.

The next good review was that of Duff and Murray in 1941 [*5*]. They included in their series only cases in which the diagnosis was confirmed at autopsy. Of their seventy-one cases, forty-eight were associated with pregnancy. Of the twenty-three that were not, nine were associated with infection. In those associated with pregnancy, the authors found that almost two-thirds of the patients were older than thirty years. Usually the pregnancies terminated in premature delivery of a stillborn child. Headaches or visual disturbances were mentioned prior to delivery in one-half of the cases. The number of previous pregnancies varied from zero to fifteen. The onset of anuria was usually on the day of delivery, but ranged from four days prior to delivery to three days following delivery. Duff and Murray pointed out that it is conceivable for bilateral cortical necrosis "to occur to an extent sufficient to cause a period of oliguria or anuria, but not sufficient to exclude the possibility of restoration of function in the non-necrotic portions of the cortex with the recovery of the patient."

Dunn in the same year (1941) reported fifteen cases of bilateral renal cortical necrosis, seven of which were associated with pregnancy [*6*]. He viewed cortical necrosis as the pathological end result of several different disease mechanisms; an inflammatory type of cortical necrosis as represented by acute necrotizing glomerulonephritis, an ischemic type associated with the toxemias of pregnancy, and a venostatic type as represented by acute renal vein thrombosis. He considered the initial event in the pathogenesis of bilateral renal cortical necrosis to be a sudden universal dilatation of the glomerular capillaries (secondary to bacterial toxins, anoxia, etc.) causing an increased intracapillary pressure which in turn forced plasma out of the capillary, thus increasing the blood viscosity. This increase in viscosity would cause a slowing and stasis of the blood flow with the resultant formation of fibrin thrombi. Anoxia would then occur secondary to the fibrin thrombi, causing a tubular necrosis. Finally, the afferent arterioles would dilate, and thrombi would be formed in backward progression to involve the interlobular arteries.

Since that time, bilateral renal cortical necrosis has been reported in the English literature as occurring in association with multiple fractures

and internal hemorrhages [7], severe burns, [8] perforated peptic ulcer [9], pregnancy (usually abruptio placentae) [10–27], influenza [28], periarteritis nodosa [29], diarrhea and dehydration [30–32], dissecting aneurysm and Marfan's disease [33], respiratory infections [34], partial heart block [35], acute appendicitis with perforation and peritonitis [36], phosphorus poisoning [37], incompatible blood transfusions [38], favism [39], a day old infant [32], and one case in which there was no discernible associated disease [40]. The first report of cortical necrosis in infants was that of Campbell and Henderson in 1949 [30]. Sheldon first noted the association of cortical necrosis with pituitary necrosis in 1942 [10]. This same association was noted by Tomlinson in 1945 [12], and then by Doniach [18] Grasby [19], MacGillivray [23] and McKay [41]. Sheldon also was the first to note ischemic necrosis of the adrenal glands in conjunction with bilateral renal cortical necrosis.

The incidence of clinical bilateral cortical necrosis is difficult to ascertain since a definite diagnosis can be made only at autopsy or, in patients who recover, by renal biopsy [40,42]. Zuelzer, in reviewing autopsy protocols on infants and children for a ten-year period, found eleven cases of cortical necrosis in 2,058 autopsies, an incidence of 0.53 per cent [32]. These eleven cases constituted one-fourth of the children dying with major renal vascular disease and led him to conclude that "cortical necrosis probably represents the commonest singular cause of renal insufficiency in infants." MacGillvray found an incidence of eight cases in 587 autopsies at a maternity hospital, an incidence of 1.4 per cent [25]. As is true with any set of statistics, the incidence varies with the samples chosen.

From the foregoing statistics, we can only conclude that, at autopsy, bilateral cortical necrosis is probably more common in pregnant women than in children. Yet, if the sample chosen included only pregnant women with bleeding in the last trimester, the incidence would be still higher and would be highest with pregnant women with accidental hemorrhage (abruptio placentae) during the last trimester. In keeping with this, Sheehan and Moore [43] found that in approximately 3.5 per cent of all patients with abruptio placentae a gross or patchy cortical necrosis will develop and will be sufficiently extensive to cause death, while in another 8 per cent a less severe grade of cortical necrosis will develop and most of these patients survive. Wahle in 1953 [38] added seventy-eight cases he gathered from the literature to the seventy-one cases previously reported by Duff and made an interesting tabulation of the combined 149 cases. Eighty-six were associated with pregnancy while sixty-three were not. Abruptio placentae alone or in combination with preeclampsia or eclampsia constituted fifty-one of eighty-six cases (60 per cent) associated with pregnancy. Of the sixty-three cases not associated with pregnancy, infection was the leading etiological agent (twenty-five cases being reported), then poisons and drugs in twelve cases, and vomiting or diarrhea in eight cases. Generally speaking, bilateral cortical necrosis is associated with abruptio placentae or toxemia in women, and infections or poisons in men. In children, it is most often associated with vomiting, diarrhea and dehydration. It is interesting to note that trauma, which is one of the leading etiological agents in acute tubular necrosis, constituted only 0.5 per cent of the cases of cortical necrosis reported by Wahle.

PATHOLOGY

Inspection of the kidneys reveals them to be somewhat swollen and soft with a capsule that strips easily, revealing a surface with a mottled appearance of red and yellow depressions. Longitudinal sectioning reveals a "spared immediately subcapsular reddish brown strip (cortex corticis) of 1 to 2 mm., beneath which the remainder of the cortex is abruptly demarcated by a yellowish grey necrotic zone extending the length of the kidneys including the cortical columns of Bertin" [44]. The greyish necrotic area of the cortex may vary from a few scattered patchy areas to a massive confluent area. The medulla is spared. On gross examination, the major renal blood vessels usually exhibit no pathologic condition.

Histologically, the small subcapsular rim of cortical tissue appears intact. This has been attributed to a collateral subcapsular circulation. Both the inner edge of this cortical rim of intact tissue and the outer edge of the medulla exhibit a border of hyperemic glomeruli. The glomerular capillaries as well as the afferent arterioles may appear markedly dilated and may contain fibrin thrombi, conglutinated red cells or fat droplets within their lumens. The necrotic tubular epithelium may exhibit poor staining qualities and nuclear degeneration. Hyaline, epithelial or

red cell casts may be present within the lumens of the tubules. The necrotic areas are surrounded by an infiltration of polymorphonuclear leukocytes and red blood cells. The small arteries, including the interlobular arteries, may have medial necrosis of their walls.

Associated lesions, when found, usually involve necrosis of the anterior pituitary gland and, infrequently, of the adrenals. Campbell and Henderson noted associated vascular lesions in the mesenteric circulation [*30*].

CLINICAL FEATURES

The dominant clinical feature is the sudden onset of gross hematuria and oliguria proceeding to an anuric state and subsequent uremic syndrome. The blood pressures are within normal limits or are slightly elevated. The patient may complain of bilateral lumbar pain or mid-epigastric pain radiating to the loins. The urine passed in the oliguric stage may be grossly bloody, may have a high specific gravity, may contain albumin and may have a sediment consistent with acute renal failure (many epithelial cell casts and epithelial cells, some red cell casts and broad renal failure casts). With the continuation of anuria, progressive acidosis, azotemia and hyperkalemia are the rule. Death occurs usually from two to thirty-two days after the onset of the anuria [*44*]. Some patients may have oliguria following anuria and then proceed to a fatal outcome. Dysuria usually is not a complaint. In contrast to acute tubular necrosis, many of these patients are anuric prior to the development of overt clinical shock. Other diagnoses often entertained in these patients are ureteral or urethral obstruction, bilateral renal infarction [*45–46*], acute glomerulonephritis, acute pyelonephritis, necrotizing papillitis, or one of the diffuse "collagen diseases." MacGillivray noted a rise in temperature on about the third oliguric day in all his eight patients with cortical necrosis whereas only one patient with acute tubular necrosis showed a similar rise [*25*]. He also noted that the blood urea nitrogen levels tend to remain lower in cases of cortical necrosis associated with pituitary infarction [*23*].

Cortical necrosis should be suspected in children with vomiting, diarrhea, dehydration and persistent oliguria, in males of any age with overwhelming sepsis (bacterial shock), and in multiparous females over the age of thirty years who have had pre-existing renal disease, or hypertension or toxemia with the present pregnancy and who present with vaginal bleeding during the last trimester of pregnancy. Of interest is the bleeding tendency in certain of these patients with abruptio placentae and its relationship, if any, to cortical necrosis. It is now appreciated that a certain number of these patients will exhibit a hypofibrinogenemia either secondary to an active fibrinolytic process or secondary to a circulating thromboplastin-like substance liberated from the bleeding uterus. This is an important consideration for the clinician since such an emergency demands immediate whole blood replacement and intravenous fibrinogen therapy prior to any surgical intervention. All patients with bleeding in the last trimester should have periodic observation of clot retraction for deficiency of clot formation or instability of a clot already formed. A fibrinogen level of oxalated blood should be obtained if possible.

Renal biopsy was employed by Gormsen, Iversen and Raaschou [*40*], who diagnosed cortical necrosis in a twenty-one year old nonpregnant woman who had sudden onset of bilateral lumbar pain. This was followed by complete anuria for five days. A tomogram taken sixty-one days after the onset of anuria showed diffuse cortical calcinosis.

EXPERIMENTAL WORK

The aim of experimental research has been to reproduce a vasospastic state of renal vessels of sufficient degree to cause an ischemic bilateral cortical necrosis. This vasospasm has been produced principally by toxins, poisons and vasoconstrictor substances. Included among the substances known to cause bilateral cortical necrosis are staphylococcal filtrate [*47,48*], hog cholera virus vaccine [*49*], intravenous lithium carmine [*50*], dioxane [*51*], diethylene glycol [*52–53*], almond extract [*54*], Bacillus coli endotoxin [*55*], meningococcal infections [*56*], horse serum combined with killed group A streptococci [*57*], pitressin® [*58*], epinephrine hydrochloride [*59*] and serotonin [*60*].

Apitz in 1934 [*55*] injected bacterial cultural filtrates intravenously into rabbits. One injection was given and repeated in twenty-four hours. These produced death in forty-eight hours with the characteristic renal lesions of bilateral cortical necrosis. He noted that this paralleled the Shwartzman skin reaction in which an initial subcutaneous injection is fol-

lowed twenty-four hours later by an intravenous injection with resultant necrosis at the injection site. Because of visceral involvement in his studies, he adopted the term "generalized" (visceral) Shwartzman phenomenon as opposed to the classical "local" (skin) Shwartzman phenomenon. He was also able to produce bilateral cortical necrosis in pregnant rabbits by a single intravenous injection of bacterial filtrates.

DeNavasquez in 1938 produced bilateral cortical necrosis by injecting staphylococcal toxin intravenously into rabbits [*48*]. He believed that the toxins principally affect the media of smaller renal arteries and that they work by causing vasoparalysis of the afferent arterioles and glomerular capillaries, followed by vasodilatation, obstruction and subsequent medial necrosis. Trueta and his colleagues [*61*] applied a tourniquet to the hind limb of a rabbit and produced spasm of the femoral and iliac arteries. They noted cortical ischemia of the kidneys. Trueta postulated that any stimulus causing a diversion of flow from the cortical nephrons to the juxtamedullary circulation would result in cortical ischemia and reflex anuria. This same shunting mechanism has also been used to explain the mechanism of renal medullary necrosis [*62*] by postulating that with the shunting mechanism in operation, circulating toxins will selectively damage the renal medulla and spare the renal cortex.

There is much evidence to refute the Trueta shunt as the mechanism responsible for cortical necrosis in man. Most of the experimental work has been carried out on rabbits which are notorious for their labile sympathetic nervous system so that renal vasoconstriction is more easily induced in them than in man. It has not been possible to carry out these same experiments successfully in dogs. Another argument against a diversion of flow is derived from a comparison of PAH extraction ratios. Man has a high extraction ratio for PAH and this extraction of PAH is believed to occur in the proximal tubules of the nephrons. The efferent arterioles of the juxtamedullary nephrons do not supply the proximal tubules, as do the efferent arterioles of the outer cortical nephrons. Therefore, when the shunting mechanism is called into action, a lower extraction ratio is to be expected. This has been found to be true of experimental shock produced in rabbits [*63*] but not in man [*64*]. Smith explains this species difference on a phylogenetic basis [*66*]. He states that in all mammals the juxtamedullary nephrons develop ahead of the cortical nephrons and that man is born with a full complement of glomeruli, whereas the white rat and possibly the rabbit double their number of glomeruli after birth. The juxtamedullary glomeruli which develop first in the white rat receive a meager sympathetic innervation, whereas the cortical glomeruli which develop at a later date receive a more effective innervation. In man, since the juxtamedullary and cortical glomeruli are all differentiated at birth, there should be no innervation difference to account for a shunting mechanism.

Tracy in 1950 was able to reproduce the Trueta shunt in anesthetized rabbits [*67*]. If deep anesthesia was used the shunting mechanism failed to operate. His experiment consisted in stimulating the renal pedicle nerves, cystoscopy with stimulation of the vesical neck and lower ureters by electrodes, and stimulation of the spinal cord by acupuncture.

Black-Schaffer, Hiebert and Kerby in 1947, in their study of purpuric meningococcemia in relation to the Shwartzman phenomenon, reproduced cortical necrosis by continuous saline infusions of meningococci into rabbits [*68*]. They were able to produce cortical necrosis without administering a priming dose intradermally and concluded that the Shwartzman reaction acts on the interlobular arteries of the kidneys producing vasospasm and ischemic necrosis.

Thal and Enger in 1955 injected staphylococci intravenously into rabbits and at various intervals of time observed the kidneys at laparotomy, at which time they would inject India ink into the abdominal aorta near the origin of the renal arteries in order to observe the renal circulation [*47*]. In many of their cases transitory renal ischemia was noted, and bilateral cortical necrosis was found in all rabbits that died. It was thought that the renal ischemia was more prolonged in them. The authors identified the injected staphylococcal toxin as the etiological agent by the fact that toxin-antitoxin mixtures failed to produce the renal ischemia. They believed that the initial disturbance was a spasm of renal venules or veins, resulting in an extreme "stagnation," hyperemia and functional ischemia. They concluded that the administered toxin, the duration of its effects, and individual variations in susceptibilities were more impor-

tant in producing the cortical necrosis than was any proposed shunting mechanism.

Cortical necrosis has been reproduced in rabbits by the intravenous injection of gram-negative endotoxins. This has been verified by several investigators. Gronvall and Brunson were successful in producing cortical necrosis in rats given gram-negative endotoxins [*69*]. They found that when gram-negative endotoxins were given in conjunction with a high molecular weight acidic polymer (liquoid®; sodium polyanetholsulfonate) intraperitoneally, or when large amounts of the acidic polymer alone were given intraperitoneally, cortical necrosis would be produced in 20 per cent of the rats tested. At postmortem, the kidney sections displayed prominent fibrinoid deposits which they believed caused the cortical necrosis by occluding the glomerular capillaries. They showed that they could minimize the severity of the renal fibrinoid lesions by the prior administration of heparin. They attributed this to some alteration of the blood coagulation mechanism related to the production of the fibrinoid deposits. They also made serial determinations of the plasma heparin-precipitable protein fraction during the course of their experiments in producing the fibrinoid lesions and found this fraction to be minimal when the fibrinoid deposits were the greatest, suggesting that this material is involved in fibrinoid production. From their work, they postulated that fibrinogen is involved in the formation of the fibrinoid renal lesions which in turn were the direct cause of cortical necrosis.

A recent paper by Page and Glendening deals with the experimental production of cortical necrosis by a vasoconstrictor substance [*60*]. They noted that the acute tubular necrosis ("shock kidney") which may follow postpartum hemorrhage is usually accompanied by extreme hypotension, while in bilateral cortical necrosis the blood pressures are generally in the normal range. The authors therefore postulated that a vasoconstrictor substance may play a part in the production of cortical necrosis. In their work they produced cortical necrosis by continuous intravenous infusions of serotonin. They explained the pathogenesis of the ischemic necrosis as follows: A retroplacental hematoma is formed in cases of abruptio placentae and this, by pressure-effects, forces some thromboplastin-like proteins from the placenta (which is one of the richest sources of thromboplastin) into the venous sinuses and from there to the maternal systemic circulation. Here coagulation takes place with patchy deposition of fibrin and resultant reduction in circulating platelets and fibrinogen. As the platelets are used up in the coagulation process, serotonin is released from them, causing a cortical vasoconstriction and subsequent ischemic cortical necrosis. In normal blood, practically all the serotonin is contained in the platelets. These investigators are now carrying out studies determining serotonin serum levels in the platelet-free blood and urine assays for serotonin metabolites in cases of abruptio placentae. No cases of carcinoid tumors have been reported with characteristic lesions of cortical necrosis.

TREATMENT

Being a variant of acute renal insufficiency, cortical necrosis deserves the same treatment as do other causes of acute renal insufficiency, such as acute tubular necrosis. General measures include careful hydration, reduction of the end-products of protein catabolism, reduction in the serum potassium load, and correction of the acidosis. The indications for dialysis are prolonged anuria, a fulminating uremic syndrome or a significant hyperkalemia, not controllable with carboxylic resins. The twenty-five and thirty-nine-day survivals reported here, together with the proved recoveries in the case of Gormsen and a case we have seen with Dr. Arthur Merrill suggest that even in this severe renal lesion, hemodialysis may have a valuable role. In "patchy" cortical necrosis, such as in Case III, the prognosis may be a little better than in acute tubular necrosis.

A new avenue of approach to the treatment of ischemic renal disease has been tried experimentally by Veghelyi and his co-workers [*70*], who lowered oxygen demands in dogs by means of chlorpromazine® and hypothermia.

SUMMARY

Bilateral renal cortical necrosis is a pathological diagnosis of a clinical entity characterized by the abrupt onset of gross hematuria, oliguria and anuria, usually in individuals without known renal disease, and proceeding to a uremic syndrome with a predominantly fatal outcome. Pathologically, there is a patchy to diffuse bilateral ischemic coagulative cortical necrosis, with characteristic sparing of the renal

medulla. Its greatest incidence is in pregnant women with accidental hemorrhage (abruptio placentae) in the last trimester. It may also occur in infants and children, usually as a result of diarrhea and dehydration, while in men it usually occurs as a result of infections or poisons. Its main clinical feature is the sudden onset of oliguria and anuria. The clinical diagnosis is difficult but is aided by percutaneous renal needle biopsy. Experimentally, it has been produced in animals principally by intravenous injection of bacterial toxins and vasoconstrictor substances. Although it is believed to result from spasm of renal interlobular arteries and afferent arterioles, its exact pathogenesis remains obscure. The treatment is similar to that employed in other causes of acute renal insufficiency. Proper management makes survival possible.

REFERENCES

1. Schreiner, G. E. Mental and personality changes of uremia. Twenty-fifth Ross Symposium. (In press.)
2. Juhel-Renoy, E. De l'anurie précoce scarlatineuse. *Arch. gén. de Méd.*, 17: 385–410, 1886.
3. Bradford, J. R. and Lawrence, T. W. P. Endarteritis of the renal arteries causing necrosis of both kidneys. *J. Path. & Bact.*, 5: 195–201, 1898.
4. Ash, J. E. Bilateral cortical necrosis of kidneys. *Am. J. M. Soc.*, 185: 71–86, 1933.
5. Duff, G. L. and Murray, E. G. D. Bilateral cortical necrosis of the kidneys. *Am. J. M. Sc.*, 201: 428–450, 1941.
6. Dunn, J. S. and Montgomery, G. L. Acute necrotizing glomerulonephritis. *J. Path. & Bact.*, 52: 1–16, 1941.
7. McFarlane, D. Focal renal cortical necrosis in a fatal case of shock. *J. Path. & Bact.*, 52: 406, 1941.
8. Brown, C. E. and Crane, G. L. Bilateral cortical necrosis following severe burns. *J. A. M. A.*, 122: 871–873, 1943.
9. Godwin, B. and McCall, A. J. Cortical necrosis complicating perforated gastric ulcer. *Lancet*, 2: 512–513, 1941.
10. Sheldon, W. M. and Hertig, A. T. Bilateral cortical necrosis; 2 cases. *Arch. Path.*, 34: 866–874, 1942.
11. Puumala, R. H. and Wells, A. H. Case, bilateral cortical necrosis. *Minnesota Med.*, 27: 375–376, 1944.
12. Tomlinson, W. J. Cortical necrosis of kidneys associated with necrosis of pituitary in obstetric shock. *Am. J. Obst. & Gynec.*, 49: 236–245, 1945.
13. Pritzker, H. G. and Rose, R. Bilateral cortical necrosis. *Canad. M. A. J.*, 52: 72–74, 1945.
14. Rinker, J. R. Postpartum bilateral cortical necrosis with recovery. *Am. J. Surg.*, 72: 286–288, 1946.
15. Clinico-pathological conference. Bilateral cortical necrosis of kidneys. *North Carolina M. J.*, 7: 316–319, 1946.
16. Atherton, H. E. Bilateral cortical necrosis in pregnancy; preliminary case report. *Memphis M. J.*, 21: 56–58, 1946.
17. Clinico-pathological conference. Symmetrical necrosis of renal cortex. *New England J. Med.*, 234: 416–420, 1946.
18. Doniach, I. and Walker, A. H. C. Combined anterior pituitary necrosis and bilateral necrosis of kidneys following concealed accidental hemorrhage. *J. Obst. & Gynec., Brit. Emp.*, 53: 140, 1946.
19. Grasby, E. D. Y. Combined anterior pituitary necrosis and symmetrical cortical necrosis of kidneys following accidental hemorrhage. *J. Obst. & Gynec., Brit. Emp.*, 54: 203–213, 1947.
20. Dodds, G. Accidental hemorrhage with bilateral cortical necrosis treated by artificial kidney. *Proc. Roy. Soc. Med.*, 41: 678–680, 1948.
21. Thomas, P. C. Renal cortical necrosis; case. *M. J. Australia*, 1: 67–68, 1949.
22. Lowrie, R. J. and O'Connell, E. B. Bilateral cortical necrosis. *New York J. Med.*, 50: 1501–1502, 1950.
23. MacGillivray, I. Combined renal and anterior pituitary necrosis. *J. Obst. & Gynec., Brit. Emp.*, 57: 924–930, 1950.
24. Smith, A. and Muirhead, E. E. Bilateral cortical necrosis of kidneys; report of 2 cases and consideration of pathogenesis. *Texas J. Med.*, 47: 88–96, 1951.
25. MacGillivray, I. Bilateral renal cortical necrosis in obstetric practice. *J. Obst. & Gynec., Brit. Emp.*, 58: 92–100, 1951.
26. Clinical pathological conference. Bilateral renal cortical necrosis in pregnancy. *J. Iowa M. Soc.*, 42: 17–23, 1952.
27. Burt, R. L. and Kearns, P. R. Bilateral cortical necrosis of the kidneys; a case report with laboratory and microscopy findings. *Obst. & Gynec.*, 2: 484–494, 1953.
28. Akerren, Y. Case of bilateral cortical necrosis of kidneys with noticeable hypochloremia. *Acta med. Scandinav.* (supp.) 196: 273–284, 1957.
29. Clinical pathological conference. Periarteritis nodosa; diffuse cortical necrosis, secondary. *New England J. Med.*, 236: 670–672, 1947.
30. Campbell, A. C. and Henderson, J. L. Symmetrical cortical necrosis of the kidneys in infancy and childhood. *Arch. Dis. Child.*, 24: 269, 1949.
31. Clinical pathological conference. Ischemic necrosis of kidneys. *New England J. Med.*, 241: 620–622, 1949.
32. Zuelzer, W. W., Kurnetz, R. and Charles, S. Circulatory diseases of the kidneys in infancy and childhood. I. Symmetrical cortical necrosis. *Am. J. Dis. Child.*, 81: 1, 2–14, 1951.
33. Lindeboom, G. A. and Bouwer, W. F. Dissecting aneurysm (and renal cortical necrosis) associated with arachnodactyly (Marfan's disease). *Cardiologia*, 15: 12–20, 1949.
34. Werner, W. A., Mucherman, R. I. C. and Allen, H. N. Bilateral cortical necrosis of kidneys. *J. Missouri M. A.*, 47: 20–22, 1950.
35. Haft, D. E. and Prior, J. T. Bilateral cortical necrosis of the kidneys following treatment of an unusual case of heart block. *Ann. Int. Med.*, 34: 6, 1483–1491, 1951.
36. McQueeney, A. J. and Speed, H. K. Bilateral cortical necrosis of the kidney. *Rocky Mt. M. J.*, 49: 513–516, 1952.

37. Perry, J. W. Phosphorus poisoning with cortical necrosis of the kidney; a report of 2 fatal cases. *Australian Ann. Med.*, 2: 94–98, 1953.
38. Sahle, G. H., Jr. and Muirhead, E. E. Bilateral renal cortical necrosis in a child associated with an incompatible blood transfusion. *Texas J. Med.*, 49: 770–775, 1953.
39. Casper, J. and Shulman, J. Bilateral cortical necrosis of the kidneys in an infant with favism. *Am. J. Clin. Path.*, 26: 42–47, 1956.
40. Gormsen, H. and Iversen, P. and Raaschou, F. Kidney biopsy in acute anuria; with a case of acute bilateral cortical necrosis. *Am. J. Med.*, 19: 209–221, 1955.
41. McKay, D. G., Merrill, S. J., Weiner, A. E., Hertig, A. T. and Reid, D. E. The pathologic anatomy of eclampsia, bilateral renal cortical necrosis, pituitary necrosis, and other acute fatal complications of pregnancy and its possible relationship to the generalized Shwartzman phenomen. *Am. J. Obst. & Gynec.*, 66: 507, 1953.
42. Crook, A. Communication on necrosis of cortex of kidney after labour. *Proc. Roy. Soc. Med.*, 20: 27–38, 1927.
43. Sheehan, H. L. and Moore, H. C. Renal Cortical Necrosis and the Kidney of Concealed Haemorrhage. Springfield, Ill., 1953. Charles C Thomas.
44. Allen, A. C. The Kidney; Medical and Surgical Diseases, p. 474. New York, 1951. Grune & Stratton.
45. Hoxie, H. J. and Coggin, C. B. Renal infarction; statistical study of 205 cases and detailed report of unusual case. *Arch. Int. Med.*, 65: 587–594, 1940.
46. Kaiser, R. F. and Ross, R. R. Total infarction of the kidneys from bilateral arterial emboli. *J. Urol.*, 66: 500, 1951.
47. Thal, A. P. and Enger, W. Selective renal vasospasm and ischemic renal necrosis produced experimentally with staphylococcal toxin. *Am. J. Path.*, 31: 233–259, 1955.
48. DeNavasquez, S. Experimental symmetrical cortical necrosis of kidneys produced by staphlococcus toxin, study of morbid anatomy and associated circulatory and biochemical changes. *J. Path. & Bact.*, 46: 47–65, 1938.
49. Rohrer, H. Kidney necrosis in acute hog cholera. *Virchows Arch. f. Path. Anat.*, 284: 203, 1932.
50. Reyna, F. G. Toxic effect of intravenous injection of lithium carmine; relation to total nephrosis of kidneys and of suprarenal cortex. *Beitr. z. path. Anat. u. z. allg. Path.*, 97: 267, 1934.
51. Barber, H. Hemorrhagic nephritis and necrosis of liver from dioxan poisoning. *Guy's Hosp. Rep.*, 84: 267, 1934.
52. Geiling, E. M. K. and Cannon, P. R. Pathological effects of elixir of sulfanilamide (diethylene glycol) poisoning; clinical and experimental correlation; final report. *J. A. M. A.*, 111: 919, 1938.
53. Berman, L. B. and Schreiner, G. E. Nephrotoxic lesion of ethylene glycol. *Ann. Int. Med.*, 46: 612–613, 1957.
54. Garvin, C. F. and van Wezel, N. Bilateral cortical nephrosis of kidney; report of 3 cases. *Arch. Int. Med.*, 62: 423, 1938.
55. Apitz, K. A study of the generalized Shwartzman phenomenon. *J. Immunol.*, 29: 255, 199, 1935.
56. Thomas, L. and Good, R. A. Studies on the generalized Shwartzman reaction. II. The production of bilateral cortical necrosis of the kidneys by a single injection of bacterial toxin in rabbits previously treated with thorotrast or trypan blue. *J. Exper. Med.*, 96: 625–641, 1952.
57. More, R. H. and Kobernick, S. D. Arteritis, carditis, glomerulonephritis, and bilateral renal cortical necrosis induced in rabbits by injection of horse serum or bovine gamma globulin combined with killed group A streptococci or Freund's adjuvant. *Arch. Path.*, 51: 361–378, 1951.
58. Bryon, F. B. Morbid effects of vasopressin on organs and vessels of rats. *J. Path. & Bact.*, 45: 1–16, 1937.
59. Penner, A. and Bernheim, A. L. Acute ischemic necrosis of kidneys; clinicopathological and experimental study. *Arch. Path.*, 30: 465–480, 1950.
60. Page, E. W. and Glendening, M. B. Production of renal cortical necrosis with serotonin (5-hydroxytryptamine), theoretical relationship to abruptio placenta. *Obst. & Gynec.*, 5: 781, 688, 1955.
61. Trueta, J., Barcly, A. E., Daniel, P., Franklin, K. J. and Pritchard, M. Studies of the Renal Circulation, p. 187. Oxford, 1947. Blackwell.
62. Robbins, E. P. and Angrist, A. Necrosis of renal papillae. *Ann. Int. Med.*, 31: 773, 1949.
63. Montague, F. E. and Wilson, F. L., Jr. Effect of epinephrine and shock on sodium para-aminohippurate extraction by the rabbit kidney. *Am. J. Physiol.*, 159: 581, 1949.
64. Breed, E. S., Maxwell, M. H. and Smith, H. W. Significance of the renal juxtamedullary circulation in man. *Am. J. Med.*, 9: 216, 1950.
65. Phillips, R. A., Dole, V. P., Hamilton, P. B., Emerson, K., Jr., Archibald, R. N. and Van Slyke, D. D. Effects of acute hemorrhagic and traumatic shock on renal function of dogs. *Am. J. Physiol.*, 145: 314, 1946.
66. Smith, H. W. The Kidney. Structure and Function in Health and Disease, p. 835. New York, 1951. Oxford University Press.
67. Tracy, E. M. Reflex renal suppression. *J. Urol.*, 64: 63–73, 1950.
68. Black-Schaffer, B., Hiebert, T. G. and Kerby, G. P. Experimental study of purpuric meningococcemia in relation to Shwartzman phenomenon with discussion of meningococcic purpura, Waterhouse-Frederichsen syndrome and bilateral renal cortical necrosis. *Arch. Path.*, 43: 28–54, 1947.
69. Gronvall, J. A. and Brunson, J. G. Generalized Shwartzman phenomenon in rats. *Arch. Path.*, 62: 324–334, 1956.
70. Veghelyi, P. V. et al. Letter to the editor. *Lancet*, 1: 581, 1956.

Radiation Nephritis*

A Clinicopathologic Correlation of Three Surviving Cases

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THE experimental production of renal lesions by exposure of the kidney to radiation was described in 1904 by Linser and Baerman [*1*]. These investigations were extended and confirmed in the laboratory animal by other workers in subsequent years [*2,3,7,8*]. The pathologic alterations described in radiated animals in which renal failure developed consisted of tubular degeneration with marked replacement by connective tissue, glomerular hyalinization and hypertrophy of the arteriolar wall. The earliest changes following radiation were vascular and glomerular engorgement and intertubular hemorrhages. Serial study disclosed progression to chronic interstitial nephritis, loss of renal function and death in uremia. Radiotherapists, however, have claimed that the kidneys were only moderately susceptible to radiation. These organs were thought to be immune to damage when ordinary doses of x-ray were employed. Despite these assertions, nephritis has been produced in man by therapeutic radiation of the abdomen and kidneys. The recent papers by Zuelzer et al. [*12*] and by Luxton [*14*] have focused attention on the importance of this problem. Previously only scattered reports on renal damage in human subjects had appeared since Domagk [*10*] first described such a case in 1927.

We have recently had the opportunity to study the renal histology and clinical course in three patients with radiation nephritis who survived the effects of this disease. In one patient nephrectomy was performed; in the remaining two, renal tissue was obtained by precutaneous renal biopsy. The biopsies were performed by one of us (S. R. C.) according to the technic of Kark and Muehrcke [*16*].

CASE REPORTS

CASE 1. D. F., a forty-nine year old white female social service worker, was found to have an adenocarcinoma of the left ovary with extension into the serosa in December, 1955. At the age of three she had had scarlet fever and questionable nephritis. However, subsequent repeated examinations of the urine and blood pressure were normal. This was corroborated in 1947 when she underwent a hysterectomy for myomata uteri. At that time her blood pressure was 120/76 mm. Hg and the urine was normal, as were the blood urea nitrogen and the hemogram.

Bilateral salpingo-oophorectomy was performed in December 1955 and this was followed by thirty-nine x-ray treatments to the abdomen over a period of fifty-one days. Radiation was distributed to four anterior and four posterior abdominal and pelvic fields, 2,500 r skin dose to each port. Each kidney was calculated to have received 1,750 r. Following this treatment she was examined at monthly intervals. No abnormalities were noted until March 21, 1956, at which time she complained of pedal edema and mild headache. On examination the blood pressure was 186/112 mm. Hg and there was pitting edema up to the mid-calf. Urinalysis revealed 3 plus albumin, a specific gravity of 1.025, 0 to 3 red blood cells and 0 to 4 white blood cells per high powered field, finely granular, epithelial and fatty casts. The hemoglobin was 10.5 gm.; red blood cells, 3.5 million per cu. mm.; white blood cells, 6,500 per cu. mm. with a normal differential count; the erythrocyte sedimentation rate was 35 mm. in the first hour. Fasting blood sugar was 91; blood urea nitrogen, 26 to 30; creatinine, 0.9 to 1.0 mg. per cent. The creatinine clearance was 49 and 54 cc./sq. meter body surface/minute. The serum chloride was 107; bicarbonate, 24.5; sodium, 142; potassium, 4.25 mEq./L. The Kahn test was negative. The urinary albumin excretion was 0.5 gm./L. The anti-streptolysin titer was 12, the serum total complement was 5.33. After treatment with reserpine and salt restriction her blood pressure stabilized at 150–160/90–100 mm. Hg with improvement of the headache and edema. The status of her renal function, anemia and urinary findings remained unchanged. Percutaneous renal biopsy was performed on the fourteenth hospital day. Other than a few scattered areas of intertubular fibrosis, the microscopic findings were limited to the glomeruli. There was hyaline thickening of the tufts and atypical nuclei

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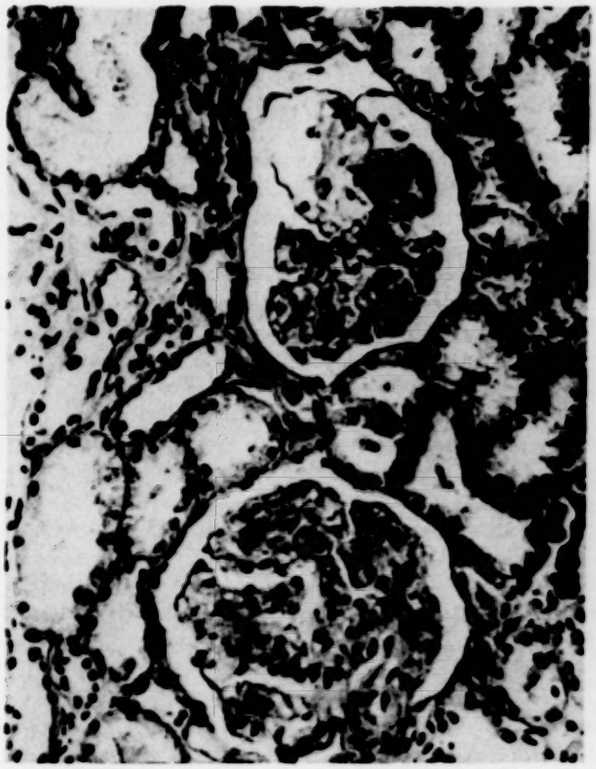

FIG. 1. This figure shows two glomeruli with hyaline thickening of the tufts and atypical nuclei indicative of cellular degeneration. There are no inflammatory cells or proliferation of Bowman's capsule. The tubules appear normal. (Case I.)

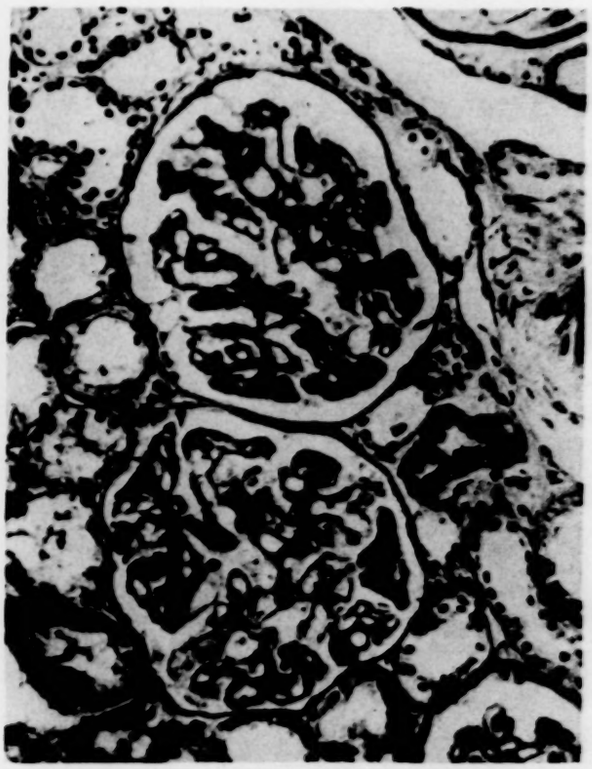

FIG. 2. In this section the major abnormalities are in the glomeruli. The tubules are normal except for scattered areas of fibrosis and thickening of basement membrane. Glomeruli show focal thickening of Bowman's capsule, thickening of tufts and atypical nuclei indicative of cellular degeneration within the glomeruli. (Case II.)

indicative of cellular degeneration. The tubules appeared normal and no inflammatory cells or proliferation of Bowman's capsule were seen. (Fig. 1.)

CASE II. Y. S., a thirty-five year old Japanese woman, had a history of rheumatic fever at the age of two. In December 1955 a papillary cystadenocarcinoma of the left ovary was removed. At the time of operation metastatic involvement of the peritoneum was noted. Physical examination prior to operation revealed a blood pressure of 120/70 mm. Hg. The examination was normal except for the ovarian tumor and the presence of a grade 3 apical, non-radiating soft systolic murmur. Fluoroscopy revealed left atrial enlargement. The fasting blood sugar was 78 mg. per cent; blood urea nitrogen, 15 mg. per cent; the hemoglobin was 11.7 gm. per cent, with a normal white blood count. The urine was free of albumin, cellular material and casts. There was no past history of renal disease. Following operation she received forty-one x-ray treatments in fifty-seven days with a total calculated dose to each kidney of approximately 2,000 r. Four anterior and four posterior abdominal and pelvic fields were radiated, with a skin dose of 2,500 r to each field. She was examined bimonthly. No abnormalities were noted until ten months following operation when she complained of morning headaches accompanied by vomiting, and weakness of six weeks' duration. At this time her blood pressure was 200/110 mm. Hg. The optic fundi, previously normal, now had an A:V ratio of 1:4, increased A-V nicking, patches of white soft exudation, and a few flame-shaped hemorrhages. She was readmitted to the hospital in October 1956. Clinical study disclosed the following: the hemoglobin was 9.2 gm., the red blood cell count, 2.9 million per cu. mm.; white blood cell count, 5,000 per cu. mm. with a normal differential count. The fasting blood sugar was 71 mg. per cent and the blood urea nitrogen was 33 to 40 mg. per cent, the creatinine ranged from 1.5 to 1.8 mg. per cent. The creatinine clearance was 25 per cent of normal. Serum sodium was 139; bicarbonate, 25; chloride, 97 mEq./L. Serum protein studies showed albumin, 3.7; globulin, 2.9; alpha-globulin, 1.0; beta-globulin, 1.0; and gamma-globulin, 1.2 gm. per cent. The cholesterol was 208 mg. per cent, with 70 per cent esters. Urinalysis made on admission revealed 1 plus albumin, 1 to 3 hyaline casts, 1 to 5 red cells and 2 to 6 white cells per high power field. Fourteen days after admission she was given reserpine 0.1 mg. four times daily. Seven days later she was given pred-

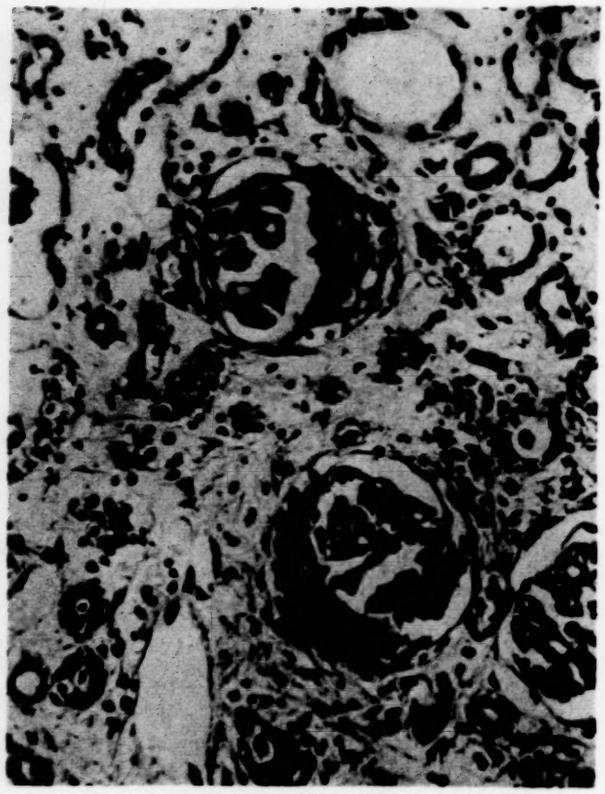

FIG. 3. This section shows a diffuse interstitial fibrosis with edema and subacute and chronic inflammation of the interstitial tissue, tubular atrophy and replacement by connective tissue. The glomeruli show marked hyalinization and necrosis of the tufts with large bizarre cells. Bowman's capsule is thickened, with crescent formation, and the blood vessels show intimal proliferation changes and thickening. (Case III.)

nisolone 40 mg. daily. By the forty-second hospital day her blood pressure ranged from 130–150 to 80–90 mm. Hg and regression of the optic findings had occurred. Repeated urinalyses disclosed albumin 1 to 3 plus, rare granular casts, many hyaline casts, 1 to 5 red cells and 2 to 12 white cells per high power field. Percutaneous renal biopsy was performed on the twenty-eighth hospital day. As in the previous section the major pathologic alterations were in the glomeruli. The tubules were normal except for scattered areas of fibrosis and thickening of the basement membrane. The glomeruli showed focal thickening of Bowman's capsule, thickening of the tufts and atypical nuclei indicative of cellular degeneration within the glomeruli. The changes in these sections appeared to be qualitatively similar to those in the previous one but were of greater magnitude. (Fig. 2.)

CASE III. F. B., a fourteen month old white boy, was discovered to have a left suprarenal neuroblastoma. At this time the urine was normal. Preoperatively he received twelve roentgen treatments in fourteen days. Three 8 by 10 cm. fields, anterior, posterior and lateral, were radiated. The dose to each field was 1,500 r skin dose. The calculated dose to the left renal bed was 2,650 r. At operation the suprarenal tumor was removed and the left kidney appeared to be normal. Following surgery he received an additional fourteen roentgen treatments in seventeen days. The total renal dose in the second series of treatments was 2,650 r; the left kidney thus received a total dose of 5,300 r over a period of thirty-six days. The child was well until three months later when he began to show irritability, anorexia and repeated emesis. He was rehospitalized. The blood pressure was 150/100 mm. Hg. The optic fundi were normal. Urinalysis disclosed 2 plus albumin, 15 to 18 red cells, 6 to 8 white cells, 3 to 4 hyaline casts, 4 to 5 granular casts, and 1 to 3 degenerated cellular casts. The hemoglobin was 11.2 gm. and the red blood cells were 4.7 million per cu. mm. The blood urea nitrogen was 22 mg. per cent and the creatinine was 0.8 mg. per cent. Serum chlorides were 84; bicarbonate, 17.3; sodium, 136; and potassium, 4.45 mEq./L. The serum albumin was 2.9; globulin, 2.4 gm. per cent. Intravenous pyelography revealed a non-functioning left kidney. Cystoscopy confirmed this, and retrograde pyelography was normal. Left nephrectomy was performed. Following surgery the blood pressure gradually subsided to 110/60 mm. Hg and the urine became normal. The child has been followed-up for one year now and is apparently well. On gross examination the excised left kidney was pale and the surface exhibited numerous pitted areas. Microscopically there was a diffuse interstitial fibrosis with edema and subacute and chronic inflammation of the interstitial tissue, tubular atrophy and replacement by connective tissue. The glomeruli showed marked hyalinization and necrosis of the tufts with large bizarre cells. Bowman's capsule was thickened, with crescent formation, and the blood vessels show intimal proliferative changes and thickening. (Fig. 3.)

COMMENT

The incidence of radiation nephritis in human subjects cannot be determined by a study of the literature. From the paucity of reports one is led to assume that this is a relatively rare occurrence, considering the large number of patients being treated with abdominal radiation. Edsall [*4*] found nitrogen retention in patients and animals exposed to abdominal radiation. In 1927 Domagk [*10*] reported the case of a nine year old child who died of renal failure following radiation of the kidneys. In 1944 Dean and Abels [*11*] observed a twenty year old woman in whom hypertension developed following renal radiation. Zuelzer [*12*] reported the deaths, in renal failure, of three children following radiation for renal tumor. Grossman [*15*] described a

similar case. Luxton [*14*] reported the development of radiation nephritis in twenty-seven of 137 patients treated for seminoma of the testis; in thirteen symptoms developed of acute nephritis such as headache, hypertension, lassitude, anemia, edema and albuminuria. Five of these patients died of renal failure. Twelve others were found to have the clinical picture of chronic glomerulonephritis when they were examined approximately one year after exposure. These patients had no symptoms of acute nephritis. The latent period for the development of acute nephritis varied from six months to one year. Two of his patients were first seen with the clinical picture of malignant hypertension. Luxton concluded that the important factor in the pathogenesis of the nephritis was the exposure of the whole of both kidneys to radiation. Kunkler [*13*] observed a similar group of men treated for seminoma in which nephritis developed in twenty-two of fifty-five patients, with seven deaths in renal failure. In 1956 Levitt and Oram [*17*] reported the case of a thirty-three year old white man in whom severe hypertension developed following abdominal radiation for seminoma of the testis. Study disclosed decreased function of the left kidney. Following left nephrectomy the hypertensive state was apparently relieved. This case is very similar to our Case III. In the latter patient we believe that a Goldblatt mechanism may have operated as a result of renal damage caused by radiation. To our knowledge no other such cases have been reported.

In our hospital these three cases have been the only instances of radiation nephritis uncovered. So far as can be estimated they did not receive a larger amount of radiation than others receiving similar therapy. Why, then, did nephritis develop in these patients? Beyond the possibilities of individual variation in susceptibility of the renal tissue to radiation and errors in dosage the remaining factors are still unknown. Luxton believes that radiation to the whole of both kidneys is an important causative factor. The British roentgenologists use more widespread radiation to the abdomen than do the American radiotherapists. This would tend to increase the cases in which the whole of both kidneys received significant amounts of radiation and may account for the apparent greater incidence of nephritis in Luxton's and Kunkler's series than that reported in this country.

An important factor which may account for the apparent infrequency of this disease entity is the lack of awareness on the part of physicians of the existence of this radiation hazard. Follow-up studies of the renal status in patients receiving abdominal radiation appear not to be made. Furthermore, as was seen in one of our cases, the relationship of the renal disease to the abdominal radiation may not be suspected and is uncovered only by chance. We believe that many cases of radiation nephritis are being overlooked. The recognition of this syndrome is of practical importance; if it is produced in significant degree by present day technics, the methods of radiation should be modified to prevent its occurrence.

Previously reported pathologic studies of radiation nephritis have been limited to tissues obtained at necropsy, and represent maximal renal damage. Luxton [*14*] and Zuelzer [*12*] described varying degrees of glomerular hyalinization, widespread fibrosis and tubular atrophy, and arteriolar fibrinoid degeneration. These findings were confirmed by Grossman [*15*].

The abnormalities in our Case 3 were similar to those described and represent end-stage tissue destruction secondary to radiation. However, the findings in our cases in which biopsies were performed were unlike those previously reported. In these the abnormalities were mainly in the glomerulus, with thickening of the tufts, degenerative cellular changes, and a few areas of intertubular fibrosis. We believe that these findings represent the more moderate stages of those previously reported, reflecting submaximal renal damage consistent with the clinical evidence of moderate renal dysfunction and survival of the patient. We consider that these changes are the earliest seen in this disease and indicate that the glomerulus is the primarily affected portion of the kidney. Zuelzer [*12*] pointed out that these findings differed from those of glomerulonephritis in the lack of inflammatory cellular infiltrate, glomerular adhesions and epithelial proliferation. We agree with this concept and add that the presence of the early degenerative glomerular changes further distinguish these two conditions and advance the concept that radiation injury primarily involves the glomerulus, with secondary tubular atrophy.

The constellation of findings described by Luxton (*vide supra*) was present in our two adult cases. No patients with radiation nephritis who survived the acute phase have been followed up over a long enough period of time to deter-

mine the prognosis of this disease. According to Luxton and Kunkler, 37 and 32 per cent, respectively, of patients in whom the acute syndrome develops die in this phase. The remainder seem to survive in the stage of chronic nephritis. Malignant hypertension may develop in a small percentage of patients. Whether or not progression continues after the development of chronic nephritis is still to be determined.

SUMMARY

1. The experimental aspects of the effect of radiation on the kidneys are briefly reviewed. A summary of the reported cases of radiation nephritis in the literature is presented and the typical clinical syndrome is summarized.

2. Three cases of radiation nephritis in which the patients survived are reported. Two adults were studied by means of renal needle biopsy. The third case was a child in whom nephrectomy was performed when a non-functioning left kidney and hypertension developed following massive radiation.

3. The pathologic study of end-stage radiation damage to the kidney is reviewed and the earlier, more mild changes are described.

4. It is suggested that the apparent rarity of this syndrome is due to a lack of awareness of its existence by the clinician and roentgenologist. It is important to establish the true incidence of this hazard of radiation therapy in order that steps may be taken to prevent its occurrence.

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REFERENCES

1. Linser, G. and Baermann, H. Ueber die lokale und allgemeine Wirkung der Roentgenstrahlen. *Fortschr. a. d. Geb. Röentgenstrahlen*, 7: 996, 1904.
2. Schulz, A. and Hoffman, B. Zur Wirkungsweise der Roentgenstrahlen. *Deutsche Ztschr. f. Chir.*, 79: 350, 1905.
3. Buschke, A. and Schmidt, H. E. Ueber die Wirkung der Roentgenstrahlen auf Drüsen. *Deutsche med. Wchnschr.*, 121: 495, 1905.
4. Edsall, D. L. and Pemberton, R. The nature of the general toxic reaction following exposure to the x-rays. *Am. J. M. Sc.*, 133: 426, 1907.
5. Heymann, P. Einwirkung der Roentgenstrahlen auf die Nieren. *Fortschr. a. d. Geb. Röentgenstrahlen*, 12: 212, 1908.
6. Hall, C. C. and Whipple, G. H. Roentgen ray intoxication: disturbances in metabolism produced by deep massive doses of the hard roentgen rays. *Am. J. M. Sc.*, 157: 453, 1919.
7. Hartman, F. W., Bolliger, A. and Daub, H. P. Experimental nephritis produced by irradiation. *Am. J. M. Sc.*, 172: 487, 1926.
8. Hartman, F. W., Bolliger, A. and Daub, H. P. Functional studies throughout the course of roentgen ray nephritis in dogs. *J. A. M. A.*, 88: 139, 1927.
9. Daub, H. P., Bolliger, A. and Hartman, F. W. The relative sensitivity of the kidney to irradiation. *Radiology*, 8: 142, 1927.
10. Domagk, G. Die Roentgenstrahlenwirkung auf Gewebe, im besonderen betrachtet an den Nieren. Morphologische und funktionelle Veränderungen. *Beitr. z. path. Anat. u. z. allg. Path.*, 77: 525, 1927.
11. Dean, A. L. and Abels, J. C. Study of the newer renal function tests in an unusual case of hypertension following irradiation of one kidney and the relief of the patient by nephrectomy. *J. Urol.*, 52: 497, 1944.
12. Zuelzer, W. W., Palmer, H. D. and Newton, W. A. Unusual glomerulonephritis in young children. Probably radiation nephritis. *Am. J. Path.*, 26: 1019, 1950.
13. Kunkler, P. B., Farr, R. F. and Luxton, R. W. The limit of renal tolerance to x-rays. *Brit. J. Radiol.*, 25: 190, 1952.
14. Luxton, R. W. Radiation nephritis. *Quart. J. Med.*, 22: 215, 1953.
15. Grossman, B. J. Radiation nephritis. *J. Pediat.*, 47: 424, 1955.
16. Kark, R. M. and Muehrcke, R. C. Biopsy of kidney in prone position. *Lancet*, 1: 1047 1954.
17. Levitt, W. M. and Oram, S. Irradiation-induced malignant hypertension: Cured by nephrectomy. *Brit. M. J.*, 2: 910, 1956.

Hypothyroidism with Anemia Demonstrating Abnormal Vitamin B_{12} Absorption*

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THE occurrence of anemia in the hypothyroid state as well as the (infrequent) association of myxedema and pernicious anemia have long been noted. It has been assumed that the uncomplicated anemia was secondary to the generalized decrease of metabolic activity, while myxedema and pernicious anemia occurring in the same patients were presumed to be pathogenetically unrelated but appeared to be therapeutically interdependent. Lack of appropriate research methods has prevented previous workers from establishing the precise nature and pathogenic mechanisms of the anemias associated with hypothyroidism. We have applied radioactive vitamins B_{12} technics in an attempt to define the pathogenesis of anemia in hypothyroidism.

REVIEW OF THE LITERATURE

The occurrence of anemia in hypothyroidism with or without frank myxedema is well documented in the literature, Charcot being the first to record the observation that patients with myxedema may be markedly anemic [*2–4*]. The type of anemia, its pathogenesis, and the relationship of pernicious anemia to myxedema have been topics of numerous case reports and discussions. Nevertheless, the basic mechanisms of anemia in the hypothyroid state remain unclear, mainly because of: (1) the limited laboratory observations in early studies (e.g. bone marrow aspirates), (2) the similarity of the clinical features and peripheral blood picture of pernicious anemia and myxedema, and (3) the only recent availability of radioactive B_{12} for the study of these anemias.

The majority of cases of anemia occurring in hypothyroidism have been classified as "uncomplicated" or "secondary" anemia [*4–7*]. There has been no unanimity of opinion regarding the classical features of this form of the anemia of hypothyroidism. The majority of case reports and experimental studies describe the anemia as macrocytic, non-megaloblastic, with or without gastric achlorhydria, and responsive to thyroid extract alone [*3,5–12*]. Such a macrocytic anemia has been observed in young rabbits after thyroidectomy and in patients following total thyroidectomy for congestive heart failure or angina pectoris [*5,10,11*]. Kunde et al. reported "rare megaloblasts" in the marrow of thyroidectomized rabbits with normal gastric acidity [*5*]. No such changes were noted in thyroidectomized patients or in the usual cases of myxedema with anemia [*11,13,14*]. Curschmann, on the other hand, found only five of 100 cases to be macrocytic ("hyperchromic") and noted many to be hypochromic [*14*]. Other authors consider the characteristic anemia to be essentially normocytic, normochromic [*15–18*]. Decourt et al. found that hypochromic anemias were predominant in their series [*13*]. However, they conclude, as does Emery, that no single type of anemia is characteristic of myxedema [*19*].

Most authors have found that the administration of iron, stomach preparations and liver was of no avail in the uncomplicated anemias [*7,17,20,21*], whereas thyroid extract ultimately was beneficial [*15,17,18*]. One case of myxedema is presented in which there was a favorable response to liver extract. However, the data are scanty, and pernicious anemia could not be definitely excluded [*22*]. Lerman and Means reported that some of their cases appeared to respond to the addition of iron and liver [*23*].

Minot studied a case of anemia in myxedema responding to thyroid extract. He interpreted the anemia (which showed no gross evidence of

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hemolysis), the leukopenia and the slight thrombocytopenia as indicative of a hypofunctioning marrow [*15*]. More extensive studies of the bone marrow in the anemia of hypothyroidism have usually shown it to be fatty and hypocellular with decreased erythropoietic activity [*5,18*]. Such bone marrow hypoplasia has been considered to be consistent with the reduction of other body functions related to the general depression of metabolism and oxidative processes [*3,4,13,15,24*].

The possibility of multiple pathogenetic mechanisms is suggested by the following facts: (1) anemia does not occur in all cases of myxedema, (2) there is no correlation between the severity of the myxedema and of the anemia, and (3) various types of anemia have been described [*3,11,25*].

The clinical similarities of pernicious anemia and myxedema, and their occasional coexistence, are of interest. Macrocytic anemia, sensations of chilliness, involvement of the tongue, sensory disturbances, edema, mental changes and achlorhydria may be found in both myxedema and pernicious anemia, and may lead to confusion in differentiation of these two conditions [*2,13,24,26*]. In 1931 Means et al. reported five cases illustrating the coexistence of pernicious anemia and myxedema, the former occurring first in three instances [*24*]. The authors did not consider that there was evidence for assuming a predisposition of either disease for the other, and each disease responded to its appropriate treatment. Leonard and coworkers reported a case of myxedema with a simple macrocytic anemia. Pernicious anemia developed while the patient was receiving thyroid therapy, and responded to the addition of liver injections [*11*]. Charaton reported the occurrence of myxedema in a case of pernicious anemia in relapse. Both conditions responded to combined therapy [*27*].

Of additional interest are several case reports in which the presence of pernicious anemia as well as of myxedema appears to be well authenticated, and in which the anemia responded only to a combination of liver and thyroid therapy [*26,28–30*]. One case of myxedema with a severe macrocytic anemia showed improvement of the myxedematous state with thyroid therapy, but only after liver extract was added did a reticulocyte response typical of megaloblastic anemia appear [*31*]. Greene describes a case of marked macrocytic anemia with achlorhydria responding to liver therapy [*26*]. Three years later myxedema with moderate anemia developed. The anemia did not respond to liver therapy before or immediately after initiation of thyroid medication, but approximately two years later the red count and hemoglobin were within the normal range. Two cases are presented by Vannotti in which megaloblastic anemia with achlorhydria appeared at the ages of twenty and twenty-two years, respectively, and was followed by myxedema [*32*]. In the first case addition of thyroid extract not only produced a response of the myxedema and anemia but made it possible to discontinue liver therapy. When thyroid was discontinued, anemia again developed.

These cases show a definite therapeutic interdependence between the two conditions. With the possible exception of the last case, however, the literature does not strongly suggest a pathogenetic relationship. Some authors have, nevertheless, speculated in this regard. Teadori believed that with the presence of achlorhydria there was a relative deficiency of an antipernicious anemia factor which could be corrected by thyroid alone [*33*]. He was of the opinion also that altered gastric function in myxedema might contribute to decreased iron absorption. Others have proposed that true pernicious anemia might develop in hypothyroidism on the basis of achylia gastrica [*24,28*]. In this regard, anemia was more severe in those patients with myxedema who also had achlorhydria [*34*]. Mansfeld demonstrated that a particular thyroid extract would support the hematopoietic activity of liver extract in thyroidectomized animals who had been made anemic by administration of saponin and collargol [*35*]. It has recently been demonstrated that there is a decreased intestinal absorption of Co^{60}-labeled vitamin B_{12} in thyroidectomized rats, and that this defect may be corrected by desiccated thyroid [*36*].

Thus, there are some tantalizing hints in the literature that the metabolism of vitamin B_{12} might be influenced by a deficiency of thyroid hormone, and that some of the cases of coexistent myxedema and megaloblastic anemia might occur on some basis other than mere chance.

METHODS

Tests for intestinal absorption of vitamin B_{12} were made according to modifications of Schilling's urinary excretion test using cobalt^{60}-labeled vitamin B_{12} [*37–39*]. Vitamin $B_{12}Co^{60}$, 1.0 or 2.0 μg. admin-

stered orally, with or without potent intrinsic factor concentrates,* was followed in zero or two hours by the parenteral injection of 1,000 μg. non-radioactive B_{12}. The twenty-four hour urine was collected and analyzed for radioactivity. Repeated parenteral injections at twenty-four and forty-eight hours, with analysis of second and third day urines, were made in a few instances [*40*].

Gastric aspirates were immediately filtered, neutralized with 0.1 N NaOH when necessary and frozen if they were not to be used immediately. Frozen specimens were thawed at room temperature overnight. Some samples were incubated at 37°c. with intrinsic factor concentrate in an attempt to demonstrate inactivation of intrinsic factor. Ten to thirty milliliters of syrup of cherry was added for flavoring immediately preceding administration.

The twenty-four hour uptake of I^{131} by the thyroid and the protein-bound I^{131} were measured by standard technics [*41*]. Thyroid stimulating hormone was given to one patient [*42*]. Other clinical studies were performed according to methods commonly employed.

CASE REPORTS

Cases Showing Abnormal $B_{12}Co^{60}$ Tests

CASE 1. This seventy-nine year old white, retired maintenance man entered the Hines Veteran Administration Hospital for the first time in October, 1955. He had been well until eight months previously. Since then he noticed a slow progression of weakness, fatigue, anorexia, weight loss of 50 pounds, lethargy, apathy, intolerance to cold, deafness and tendency to develop swelling of the ankles.

On physical examination, he was well developed, afebrile, pale and in no acute distress. Pulse was 60/minute; respirations, 24/minute; blood pressure, 130/60 mm. Hg. His skin was pale, coarse and scaly. Axillary hair was sparse, and eyebrows were thinned laterally. The tongue was beefy and enlarged, but papillary atrophy was absent. There were crepitant rales in both lung bases. The heart was not enlarged, and there were no murmurs. An umbilical hernia was present. There was moderate pitting edema of the ankles. The rest of the physical examination was non-contributory.

Several basal metabolic rates at time of admission were −20, −25 and −21 per cent. An I^{131} uptake was 4.6 per cent with .009 microcuries/L. protein-bound I^{131} (e.g. values are well within the hypothyroid range). I^{131} uptake after injection of thyroid stimulating hormone did not reveal any significant change. The blood cholesterol was 234 mg./100 ml. An x-ray of the skull showed no abnormality of the cranial bones or sella turcica.

The initial red cell count was 2.3 million/cu. mm.; hemoglobin, below 7 gm./100 ml.; hematocrit, 18 per cent; white cell count, 7,000/cm. mm.; and the blood smear, 78 per cent polymorphonuclears and 22 per cent lymphocytes. The blood smear showed moderate anisocytosis and poikilocytosis, slight polychromasia and slight targeting. The platelets were adequate, and white cells showed slight toxic granulation. The bone marrow was slightly hypercellular and contained adequate megakaryocytes. The myeloid:erythroid ratio was approximately 2:1. Erythropoiesis was predominantly of the normoblastic type, although some cells were megaloblastic. There was a slight shift to the left in the granulocytic series, with a number of giant metamyelocytes which had the bizarre configuration characteristic of megaloblastic anemia. (Figs. 1A and B.) Free acid was present in the gastric juice. Gastroscopy revealed a normal gastric mucosa. $B_{12}Co^{60}$ tests are noted in Tables I and II and under "results."

Because of the normal $B_{12}Co^{60}$ tests various studies were undertaken in an attempt to assess gastrointestinal absorption, renal function and liver function. An oral glucose tolerance curve revealed a fasting blood sugar of 111 mg./100 ml., a rise to 162 at thirty minutes, 147 at two hours and 123 at three hours. Absorption of I^{131}-tagged total fats and fatty acids was just below normal limits. X-rays of the upper gastrointestinal tract and small bowel revealed only prolapse of the gastric mucosa into the base of the duodenal bulb, with some prominent mucosal folds in the antrum, and some hypomotility of the small bowel. No abnormality was noted in the large bowel on examination by barium enema.

On admission a routine urine specimen revealed a specific gravity of 1.014, a trace of albumin, 30 leukocytes per high power field, and no casts. Urea clearance was 46 per cent of normal; phenolsulfonphthalein excretion was 10 per cent in fifteen minutes. Blood urea nitrogen values were 21.6 to 37.6 mg./100 ml. A liver "profile" including the van den Bergh test, two-hour Watson test for urine urobilinogen, thymol turbidity, serum cholesterol esters, and cephalin flocculation test were all within normal limits. However, a bromsulfalein test showed dye retention of 10 per cent at forty-five minutes.

Electrocardiogram demonstrated a first degree A-V block with a right bundle branch block. An x-ray of the chest taken on admission revealed a depression of the minor fissure and accentuation of markings in the lower right lung field consistent with a previous inflammatory condition. The heart did not appear enlarged, but there was some tortuosity of the aorta. A serologic test for syphilis was negative.

Figure 2 reveals the subsequent course of hematologic values, basal metabolic rates and their relation to therapy. Early in the course he was given two transfusions. Because of his age and the electrocardiographic abnormalities, thyroid therapy was given very

* Various lots of intrinsic factor concentrate purified from hog stomach mucosa have been obtained from Armour Laboratories.

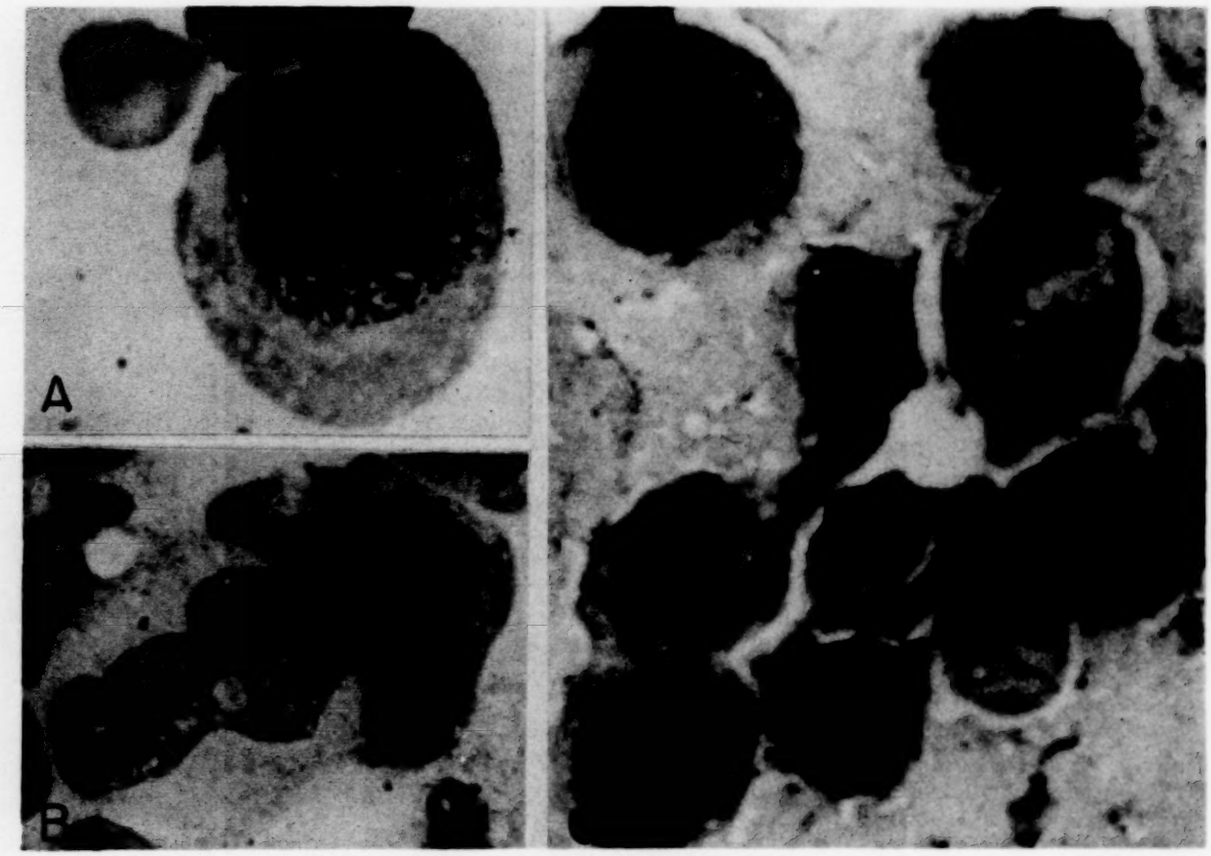

FIG. 1. A and B, bone marrow smear taken from Case I before thyroid or vitamin B_{12} therapy. A, Basophilic megaloblast. B, Orthochromic megaloblast and giant metamyelocyte. C, bone marrow smear taken from Case III during myxedematous stage, with slight anemia. Normoblasts and a normal metamyelocyte are seen. Magnification is approximately × 2,000 in each.

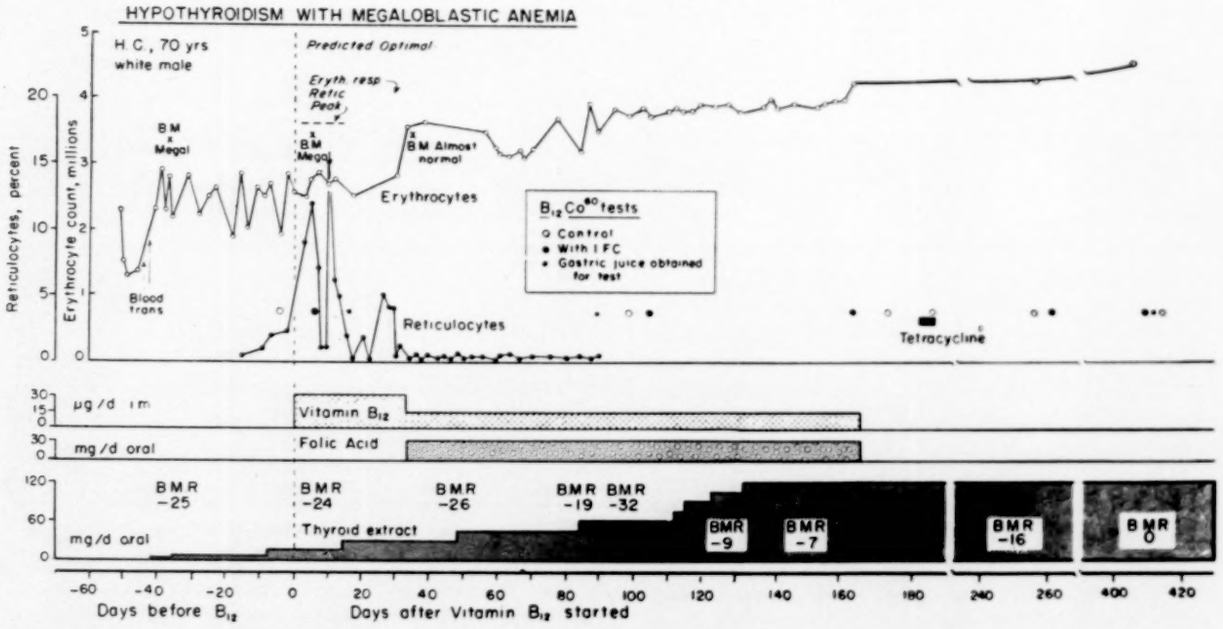

FIG. 2. Clinical course of Case I who initially presented with myxedema and megaloblastic anemia.

cautiously. He showed progressive increase in mental awareness, physical strength and basal pulse rate. The skin gradually became more normal. Parenteral vitamin B_{12} was started while the patient was receiving 15 mg. thyroid extract daily. As noted in Figure 2, the reticulocyte response reached 15 per cent but the red cell response was much slower than would usually be seen in pernicious anemia. The most recent study, more than a year after initiation of vitamin B_{12} therapy, showed a red cell count of 4.5 million/cu. mm.; hemoglobin, 13 gm./100 ml.; and hematocrit, 39 per cent. Urea clearance was 102 per cent of normal; phenolsulfonphthalein was 20 per cent in fifteen minutes. Urea nitrogen was 14.6 mg./100 ml.

Case II. This forty-six year old white railroad conductor entered Hines Veterans Administration Hospital in May, 1931, complaining of numbness in his hands and legs, anorexia, weight loss, weakness and mental apathy, all of one month's duration. He was undernourished, listless, dull and appeared chronically ill. The pulse was 76/minute, blood pressure, 108/70 mm. Hg. He walked with a slightly widened base; the Romberg test was slightly positive. There was some loss of muscular power in all extremities. There was some impairment of position sense in the lower extremities but pain and temperature sensation were intact. The remainder of the physical examination was within normal limits. The red cell count was 3.25 million/cu. mm. and the hemoglobin was 80 per cent. The white count was 8,500/cu. mm. with a normal differential. A fractional analysis of gastric juice revealed the absence of free hydrochloric acid. The blood serology for syphilis was negative.

A diagnosis of pernicious anemia with incipient involvement of the spinal cord was made. The patient was treated with daily liver extract parenterally. In two months the red cell count was 4.33 million/cu. mm. with a hemoglobin of 85 per cent, but improvement in his neurological condition was not apparent.

He failed to take liver injections as directed after release from the hospital. He was hospitalized for the second time in December, 1932, because of extreme weakness, numbness of the entire body and tingling of the hands and feet. Physical examination revealed an emaciated, anemic and chronically ill patient. The weight was 110 pounds, pulse was 84/minute, and blood pressure was 104/68 mm. Hg. There was a lemon tinge to the skin, and his hair was prematurely gray. The tongue was smooth, thin and flabby. Neurological examination now revealed difficulty in locomotion, marked diminution of power in the lower extremities and slightly exaggerated deep tendon reflexes. Position, pain and vibratory sensations were impaired, particularly in the lower extremities. The Romberg test was positive, and coordinate movements were poorly performed. The red cell count was 2.89 million/cu. mm., and the hemoglobin was 70 per cent. There was slight anisocytosis and poikilocytosis. Acid was again demonstrated to be absent from the gastric juice. Two basal metabolic rates were −14 per cent and −2 per cent. The chest x-ray was interpreted as showing chronic bronchitis. An electrocardiogram was normal. The patient was again given liver extract. Several months later, while receiving regular liver injections, he showed a red cell count of 5.4 million/cu. mm. and a hemoglobin of 90 per cent. He still complained of generalized weakness, and was unable to walk any great distance.

The patient continued to take liver injections, but only sporadically. He was admitted for the third time in June, 1940, because of a possible coronary occlusion. This diagnosis was not supported by physical and laboratory findings. The red count was 4.2 million/cu. mm. with a hemoglobin of 80 per cent.

In May, 1956, at age seventy-two, he entered for the fourth time complaining of increased weakness, inability to walk without aid, rectal incontinence, orthopnea and dyspnea of sixteen months' duration. He had been taking liver injections regularly. He had been operated on one year previously for varicocele and left inguinal hernia. Two months previously he had been treated for a viral pneumonia with pleural effusion, at which time he was found to have a diaphragmatic hernia. He had experienced several grand mal seizures prior to admission. Physical examination revealed a weak, elderly man with a sallow complexion. He had coarse, dry skin, loss of eyebrows laterally and puffiness of the lower eyelids. He was more alert and active than is usual for patients with marked hypothyroidism, however. His weight was 118 pounds, pulse was 52/minute and blood pressure was 140/90 mm. Hg. The right lobe of the thyroid gland was hard. There was dullness with crepitant, inspiratory rales in both lung bases posteriorly; otherwise the lungs were hyperresonant. The heart was enlarged to the left with the left cardiac border percussed in the anterior axillary line. There was a grade 2 systolic blowing murmur heard best at the apex. A varicocele was palpated in the left scrotal sac. Neurological examination was consistent with subacute combined degeneration of the spinal cord.

The red cell count was 3.45 million/cu. mm. with a hemoglobin of 9 gm./100 ml. The white cell count was 10,200/cu. mm. with a normal differential. There was anisocytosis and poikilocytosis. The white cells contained toxic granulation and the platelets were adequate. The reticulocyte count was 0.1 per cent. The bone marrow was normally cellular containing adequate megakaryocytes. The myeloid:erythroid ratio was approximately 2:1. Erythropoiesis was normoblastic. There were some toxic changes in progranulocytes, an increased number of eosinophils and a slight increase in plasma cells. Gastric analysis after histamine administration failed to reveal any free hydrochloric acid. Gastroscopy showed a patchy,

TABLE I

URINARY EXCRETION OF $B_{12}CO^{60}$

Case No.	Vitamin B_{12} Therapy		Thyroid Therapy		$B_{12}Co^{60}$ Urinary Excretion Tests								
						Control Tests			Intrinsic Factor Tests				
						Per cent Excreted			Intrinsic Factor Preparation		Per cent Excreted		
	Months	RBC × 10^6/cu. mm.	Months	Mg./Day	μg. $B_{12}Co^{60}$	1 st 24 hr.	2nd 24 hr.	3rd 24 hr.	Lot*	Mg.	1st 24 hr.	2nd 24 hr.	3rd 24 hr.
1	0	2.55	1	15	2	0.5	.....	.....	A	150	1.6	.....	.....
	3	3.81	4	60	1	1.8	.....	.....	B	50	2.7	.....	.....
	5	4.19	6	120	1	3.5	.....	.....	B	25	5.2	.....	.....
	5	4.19	6	120	1	3.1†	.....	.....	.....	.....	.....	.....	.....
	8	4.29	9	120	1	4.4	4.8	1.7	B	50	1.7	2.3	1.2
	None	4.50	15	120	1	5.8	.....	.....	B	25	9.0	.....	.....
2	0	3.60	0	...	1	2.2	.....	.....	B	50	1.3	.....	.....
	1	3.77	Start	7	1	.....	.....	.....	B	50	1.1‡	.....	.....
	3	3.92	2	120	1	0.1	0.0	0.0	B	50	0.1	0.0	0.0
	6	4.40	5	120	1	.....	.....	.....	B	50	3.3	.....	.....
3	None	3.77	0	...	1	2.6	.....	.....	B	50	4.1	.....	.....
4	None	4.29	7	60	1	8.8	.....	.....	.....	...	.....	.....	.....
5	None	2.58	0	...	1	16.6	.....	.....	.....	...	.....	.....	.....
6	None	3.78	0	...	1	26.4	.....	.....	.....	...	.....	.....	.....
7	None	3.90	0	...	1	12.6	.....	.....	.....	...	.....	.....	.....
Expected Values Normal					1	8–25	About ⅓	About ⅓	.....	...	.....	.....	.....
					2	6–20	of 1st	of 2nd					
Pernicious Anemia					1	0–4	24 hr.	24 hr.	1–3 U.S.P.	...	8–25	About ⅓	About ⅓
					2	0–3			units		6–20	of 1st 24 hr.	of 2nd 24 hr.

* Armour Laboratories intrinsic factor preparations: A = lot 720-059-1 (1-A); 1 U.S.P. unit is about 50 mg.
B = pool 23; 1 U.S.P. unit is about 35 mg.

† Tested on fifth day of tetracycline, 250 mg., four times daily.

‡ Tested on fifth day of chlortetracycline, 250 mg., four times daily.

atrophic gastritis which was diffusely located throughout the stomach.

Oral I^{131} uptake was 4.9 per cent at twenty-four hours, with .016 microcuries/L. protein-bound I^{131}. After administration of thyroid stimulating hormone the uptake of oral I^{131} was 2.6 per cent with .010 microcuries/L. protein-bound I^{131}. Of 50 microcuries of I^{131} given intravenously, 4.8 per cent was taken up by the gland in twenty-four hours. The serum cholesterol was 290 mg./100 ml. X-rays of the skull revealed a normal calvarium and sella turcica. X-rays of the neck revealed a calcified adenoma in the right lobe of the thyroid. Attempts to measure the basal metabolic rate were unsatisfactory.

$B_{12}Co^{60}$ studies are noted in Tables I and II, and under "results." Because of malabsorption type $B_{12}Co^{60}$ tests, a search was made for other gastrointestinal or renal defects. An oral glucose tolerance curve was within normal limits. Absorption of I^{131}-tagged fats and fatty acids was normal. X-rays of the upper gastrointestinal tract and small bowel demonstrated some decrease in prominence of the mucosal folds in the fundus of the stomach, but were otherwise normal. Three stools for occult blood were negative. On admission the urine revealed a faint trace of albumin with a few leukocytes and many bacteria. These findings were subsequently absent. Urea clearance values were 36 and 55 per cent of normal; phenolsulfonphthalein excretion was 12 to 22 per cent in fifteen minutes. Urea nitrogen values were 21.1 to 29.2 mg./100 ml.

A slight cardiomegaly and emphysema were evident on x-ray of the chest. Other laboratory studies which were normal included serologic test for syphilis, electrocardiograms, serum bilirubin and urinary urobilinogen. Spinal fluid was normal except for a non-specific colloidal gold pattern.

Seizures were attributed by the neurologists to cerebral arteriosclerosis, and responded well to dilantin.[8] He was continued on a regimen of weekly injections of refined liver extract, and was started on thyroid extract, ultimately reaching a maintenance dosage of 120 mg./day. Although he still complained of weakness and paresthesias, he noted a marked improvement in mental alertness, and was able to carry on daily activities with less difficulty. The skin gradually returned to a normal texture. Nine months after the fourth admission the red cell count was 4.4 million/cu. mm., hemoglobin was 13 gm./100 ml. and white count was 5,850/cu. mm. with a normal dif-

TABLE II

GASTRIC ASPIRATES FROM CASES I AND II TESTED FOR INTRINSIC FACTOR ACTIVITY OR INHIBITION IN CASES OF PERNICIOUS ANEMIA

Gastric Juice Donor		Pernicious Anemia Patient							Gastric Juice Test		Test of Gastric Juice Incubated with Intrinsic Factor Concentrate					
		All Tests			% 24 hr. Excretion Control Test	Intrinsic Factor Test					State of Juice	Ml.	Intrinsic Factor		Min. at 37°C.	24 hr. Excretion
Case	Time*	Patient	μg. $B_{12}Co^{58}$	24 hr. Period		Lot†	Mg.	% 24 hr. Excretion	Ml.	% 24 hr. Excretion			Lot†	Mg.		
1	1	V. R.	2	1st	*0.1*	C	125	*8.0*	25	*2.1*	.....	..	..	..	...	....
	4	M. B.	1	1st	*2.6*	B	50	*20.9*	75	*4.9*	Frozen	62	B	50	15	*13.1*
	5	M. H.	1	1st	*0.2*	B	25	*7.3*	....	.....	Fresh	20	B	25	60	*2.8*
	6	M. S.	1	1st	*1.8*	B	25	*7.4*	....	.....	Fresh	20	B	25	40	*11.2*
	15	F. H.	1	1st	*2.2*	B	15	*13.5*	98	*6.4*	.....	..	..	..	...	....
2	2	E. Z.	1	1st	*2.7*	B	15	*3.9/9.0*‡	30	*0.7*	Fresh	19	B	15	45	*8.5*
	..	.....	..	2nd	....	..	...	*1.1/2.5*	....	*0.0*	.....	..	..	..	...	*2.5*
	..	.....	..	3rd	....	..	...	*0.0/0.9*	....	*0.4*	.....	..	..	..	...	*0.1*
	5	F. H.	1	1st	*2.2*	B	15	*13.5*	....	.....	Frozen	45	B	15	115	*12.7*
Expected Response in Patients With Pernicious Anemia			1	1st	*0-4*	B	15	*5.5+*	25§	*7.0+*						
							25	*7.0+*	50	*8.0+*						
							50	*8.0+*	100	*8.0+*						

* Month of thyroid therapy when gastric juice aspirated.
† Intrinsic factor preparations from Armour Laboratories. B = pool 23; 1 U.S.P. unit is approximately 35 mg. C = 441-254; 1 U.S.P. unit is approximately 75 mg.
‡ Intrinsic factor test was repeated after all other tests completed.
§ Assuming normal donors of gastric juice.

ferential count. A urine specimen had a specific gravity of 1.014 with a few leukocytes. The urea clearance was 71 per cent.

CASE III. This retired white laborer was first studied at the Presbyterian Hospital, Chicago, seven years previously, at age sixty-seven, with classical symptoms and manifestations of myxedema, including a basal metabolic rate of −27 per cent. He was improved on thyroid medication, but stopped taking it after about two years. He had a relapse including constipation, myxedema, dry skin, cold intolerance, deepening of voice, and slowing of physical and mental processes. History revealed an episode of syphilis adequately treated with arsenicals many years previously, and chronic alcoholism with intake of 2 pints of whiskey daily over a forty year period up until the onset of relapse. Findings consistent with myxedema included dry hair and skin, large tongue, deep voice, slow reactions, edema of face and neck, and slow relaxation of deep tendon reflexes. The thyroid was not enlarged. Incidental physical findings included obesity, emphysema, bilateral hydrocoele and blood pressure of 140/90 mm. Hg. Vibration and position senses were normal. The basal metabolic rate was −29 per cent; serum cholesterol, 398 mg./100 ml.; and I^{131} twenty-four-hour uptake, 1 per cent. An electrocardiogram showed low voltage in all leads. The sella turcica was normal on x-ray. Hematologic data indicated: hemoglobin, 10.2 gm./100 ml.; hematocrit, 32 per cent; red count, 3.23 million/cu. mm.; white count, 3,000 to 5,500/cu. mm.; differential count, normal; and reticulocytes, 1.6 per cent. Liver extract, equivalent to 20 μg. B_{12} daily, was given for two weeks with no improvement in reticulocyte or red blood counts. There was no free acid in the stomach after an Ewald test meal. The cephalin flocculation test was ++++ in twenty-four hours; two-stage prothrombin time, 76 per cent of normal activity; and oral and intravenous glucose tolerance tests, normal. Urea clearance values were 45 to 54 per cent of normal; bromsulfonphthalein was 40 per cent in fifteen minutes. Blood urea N was 16 mg./100 ml. The serum albumin and globulin were normal. X-rays of the upper and lower gastrointestinal tract and proctoscopy were normal despite the presence of a trace to ++++ benzidine reactions of the stools.

He was given thyroid therapy again, with general clinical improvement. Several years later, however, he again stopped taking thyroid and again had a symptomatic relapse for which he was hospitalized. Symptoms and physical findings were essentially the same as on the previous admission. He had taken no thyroid for over a year. Laboratory studies showed basal metabolic rate −38 per cent; serum cholesterol, 411 mg./100 ml.; protein-bound iodine, 1.2 μg./100 ml. (N = 3.5–8.0); and low voltage on electrocardiogram. Blood studies showed hemoglobin, 12.4 gm./100 ml.; red count, 3.77 million/cu. mm.; hematocrit, 34 per cent; white blood count, 4,100/cu. mm.; platelets, adequate; and reticulocytes, 1.2 per cent. Differential count showed 29 per cent segmented

neutrophils, 8 per cent stab neutrophils; 50 per cent lymphocytes, 3 per cent monocytes and 10 per cent eosinophils. The bone marrow showed some decrease in total cellularity. The myeloid:erythroid ratio was about 2:1. Normal types of maturation were present. (Fig. 1C.) The serum glucose was 99 mg./100 ml. and uric acid was 4.6 mg./100 ml. Non-nitrogen nitrogen was 35 mg./100 ml. $B_{12}Co^{60}$ tests are given in Table 1 and under "results."

Thyroid therapy was resumed and the patient was discharged from the hospital. Unfortunately, he has failed to keep his return appointments, and no further studies have been possible.

Cases Showing Normal $B_{12}Co^{60}$ Tests
Results of $B_{12}Co^{60}$ tests are shown in Table 1

CASE IV.* A sixty-seven year old white man was first seen two years previously with typical myxedema including low voltage on the electrocardiogram, I^{131} uptake of 3.1 per cent, and protein-bound I^{131} of 0.063 microcuries/L. The red count was 4.29 million and hemoglobin, 14.0 gm./100 ml. He had been receiving up to 60 mg./day thyroid for seven months, and had no anemia at the time of testing.

CASE V.† A twenty-one year old white female cretin had received no thyroid therapy for ten years. Twenty-four hour I^{131} uptake was 2.1 per cent. An anemia with hemoglobin of 7.0 gm./100 ml., red count of 2.58 million/cu. mm., and mean corpuscular hemoglobin of 27 did not respond to thyroid alone but later responded promptly to iron therapy.

CASE VI.* A sixty-four year old white man had heavy neck radiation for carcinoma of the larynx two years previously, and apparently hypothyroidism developed as a result. He showed typical clinical manifestations of myxedema including I^{131} twenty-four uptake of 4.3 per cent. The red count was 3.78 million/cu. mm.; hemoglobin, 12.0 gm./100 ml.; and hematocrit, 36 per cent.

CASE VII.‡ A fifty-five year old white man had classical symptoms of myxedema for four or five years. Laboratory studies were all consistent with this diagnosis, including twenty-four hour I_{131} uptake of 2.0 per cent. The hemoglobin was 10.7 gm. 100 ml.; hematocrit, 32 per cent; and red blood cell count, 3.9 million/cu. mm.

RESULTS

Tables 1 and 11 report the results of all $B_{12}Co^{60}$ tests in these patients. In Case 1, the patient who apparently had simultaneous onset of myxedema and megaloblastic anemia with normal gastric acidity and normal gastroscopic appearance, initially showed very low excretions, both with and without intrinsic factor. This was not corrected by tetracycline administration. After nine months of thyroid therapy an intermediate range excretion of $B_{12}Co^{60}$ was observed during the first twenty-four hours, with a slightly greater excretion on the second day. The intrinsic factor test at this time was paradoxically lower. Six months later the control test was elevated slightly more but was still within the intermediate zone. The intrinsic factor test, on the other hand, had returned to normal. Two tests during the early phase of therapy, using up to 75 mg. of gastric juice, showed no significant evidence of intrinsic factor activity in cases of pernicious anemia. A test using 98 ml. of gastric juice after fifteen months of thyroid medication showed slight intrinsic factor activity. Of three tests for inhibition of intrinsic factor activity by the patient's gastric juice, one appeared to show inhibition, the other two did not.

In the second case, pernicious anemia appeared to be present for many years before hypothyroidism became evident. Both control and intrinsic factor tests were in the low range, even after therapy with chlortetracycline and after five months of thyroid therapy. The gastric juice showed no intrinsic factor activity and no tendency to inhibit intrinsic factor on two tests.

The third case of classical myxedema with achlorhydria and a normocytic anemia, could be tested only once. The patient showed poor absorption of $B_{12}Co^{60}$ both with and without intrinsic factor concentrates.

Four other patients with hypothyroidism, only one of whom was receiving thyroid therapy, had normal control responses with vitamin $B_{12}Co^{60}$. Some anemia was present in three, and one patient had classical cretinism.

COMMENTS

Three features of this study warrant discussion: (1) mechanisms responsible for the abnormal $B_{12}Co^{60}$ tests noted in three cases, (2) characterization of the anemias observed, and (3) pathophysiology of the anemia in hypothyroidism.

Abnormal $B_{12}Co^{60}$ Findings. A case of myxedema with megaloblastic anemia which we first observed in October, 1955, stimulated us to study $B_{12}Co^{60}$ absorption in seven cases of hypothyroidism, including the original case. We

* Veterans Administration Hospital, Hines, Illinois.
† Presbyterian Hospital, Chicago, Illinois.
‡ Illinois Research and Educational Hospitals, Chicago, Illinois.

demonstrated an apparent decrease of absorption in three cases, and know of a similar unreported case studied by others [*43*]. This abnormality was not corrected by the simultaneous administration of intrinsic factor, as would occur in classical pernicious anemia. Such a defect of vitamin B_{12} absorption not corrected by the addition of intrinsic factor is typical of the sprue syndrome, in which other substances are also poorly absorbed. Other studies, including complete roentgen examination of the gastrointestinal tract, oral glucose tolerance tests and radioactive fat absorption, were performed in the first two cases in a search for intestinal disease or malfunction. Apart from an equivocal decrease of fat absorption in one case, no abnormalities were demonstrated.

Three explanations other than defective intestinal absorptive ability may be postulated for a decreased urinary excretion test both with and without intrinsic factor: (1) chemical or bacterial impairment of vitamin B_{12} utilization (2) impaired renal function, and (3) inhibition of intrinsic factor activity.

Wolff and Weiss demonstrated that the destructive action of cysteine in respect to vitamin B_{12} might be prevented by the B_{12}-binding activity of gastric juice [*44*]. The binding ability of gastric juice in Case I was tested and was found to be within the normal range. Broad-spectrum antibiotics have corrected the deficient absorption of B_{12} in some cases of megaloblastic anemia related to intestinal stricture or blind pouch [*45–47*]. Presumably this indicated competition for vitamin B_{12} by bacterial flora or destruction of vitamin B_{12} by the bacteria. However, we were unable to improve the B_{12} absorption in two of our cases using the prescribed course of antibiotics. Destruction of vitamin B_{12} by other agents, however, cannot be ruled out.

Recently Rath et al. pointed out that the presence of renal disease may impair vitamin $B_{12}Co^{60}$ excretion and that intrinsic factor does not alter this impairment [*48*]. These authors failed to find any striking correlation of blood urea nitrogen levels or urinary volume with vitamin $B_{12}Co^{60}$ excretion. Their patients excreted considerable amounts of radioactive B_{12} after the first twenty-four hours, in contrast to patients with pernicious anemia. Impairment of function in all three of our patients appears slight, particularly as evidenced by the urea clearance. These slight changes are consistent with the alterations in renal dynamics in myxedema noted by others [*49,50*]. Case II showed a moderately decreased urea clearance on one occasion, but the $B_{12}Co^{60}$ tests remained abnormal, even when renal functions had returned to normal. In two tests in which successive twenty-four hour urine samples were collected, a larger amount of vitamin $B_{12}Co^{60}$ was excreted during the second twenty-four hours than during the first twenty-four hours in Case I. The reason for this is not readily apparent. Such a lag in excretion was not noted in Case II. The defective excretion of $B_{12}Co^{60}$ is apparently related to excretory functions rather than to intestinal absorption. Renal disease has never been known to cause megaloblastic anemia. Two of our patients on the other hand, had a megaloblastic anemia at some time. All these considerations minimize renal impairment as an important factor in the genesis of abnormal vitamin B_{12} excretion in our patients. We have not measured fecal excretion [*51*], blood level [*52*] or hepatic uptake [*53*] of $B_{12}Co^{60}$ in these cases, procedures which might resolve this possibility even more definitely.

A temporary decrease in urinary excretion of $B_{12}Co^{60}$ has also been noted during acute bacterial infections, but there is no reason to suspect this factor in the present cases [*54*].

We attempted to demonstrate inhibition of intrinsic factor by the gastric juice in Cases I and II on several occasions. The inconstant demonstration of apparent inhibition in Case I, and the failure to demonstrate inhibition in Case II make such a mechanism improbable. The only partial improvement on control tests, with complete return to normal of intrinsic factor tests in Case I, also speak against this postulate.

On reviewing the foregoing hypotheses for the abnormal $B_{12}Co^{60}$ tests noted in our cases, we conclude that the most likely mechanism is decreased ability of the intestine to absorb vitamin B_{12}. In addition, a decreased excretion of intrinsic factor appears evident in the first two cases, although this defect in the second case may well be coincidental. The possible relationship of these abnormalities in physiology to the hypothyroid state will be discussed subsequently. It is of interest that a recent study has shown low intrinsic factor activity of gastric juice in all patients studied who had intestinal malabsorption, regardless of the cause [*55*].

Definition of the Anemias. In Case I the patient first presented with myxedema and megalo-

blastic anemia. The presence of free acid in his gastric juice and the normal gastric mucosa noted on gastroscopy are not consistent with our usual concepts of pernicious anemia in the adult. However, presence of free acid in the stomach has been reported in five of six children with pernicious anemia [*56*]. We have also found this in one of two children with pernicious anemia whom we have tested. Thus achlorhydria, while almost always present, does not appear to be a *sine qua non* of early pernicious anemia. Wintrobe has emphasized that a lack of intrinsic factor in the gastric juice must be demonstrated before a case of megaloblastic anemia with free hydrochloric acid can be considered to be pernicious anemia [*57*]. It is our opinion that, in addition, the absence of intrinsic factor should be irreversible, and should not be secondary to other disease or surgery before a case be considered one of classical Addisonian pernicious anemia. On two early tests this patient's gastric juice showed no intrinsic factor activity, whereas after fifteen months of thyroid therapy weak activity was demonstrated. One of three tests for inhibition of intrinsic factor appeared to show such inhibition. Failure to demonstrate inhibition consistently might be explained by an inhibitory substance originating in the intestine which was inconstantly regurgitated into the stomach. If this were the case it should continue to inhibit both control and intrinsic factor tests, whereas the most recent test showed good excretion after intrinsic factor.

Although one is always treading on uncertain ground when *post hoc* reasoning is applied, particularly when a single case is concerned, still the total observations in this case are so discordant with our observations in other cases of the various megaloblastic anemias with no thyroid deficiency that we shall postulate the following mechanisms: (1) The myxedematous state in some manner led to a local intestinal defect in absorption of vitamin B_{12}. This defect returned to normal on thyroid therapy, but only after many months. (2) The myxedematous state also led to a marked decrease of intrinsic factor excretion by the stomach. This returned only partially towards normal after many months of thyroid therapy. Either this was observed too early in the disease for the development of achlorhydria and gastroscopic changes [*58*], or else this particular cause for intrinsic factor deficiency need not have such concomitants. Assuming that the intrinsic factor defect will prove to be completely reversible, we might term this first case a "reversible pernicious anemia" secondary to myxedema and showing malabsorption of vitamin B_{12}.

In Case II, when first seen by us, the patient presented clinical and laboratory evidence of myxedema and had a moderate normocytic normochromic anemia. In addition, his past history and presenting physical findings supported the diagnosis of pernicious anemia with subacute combined degeneration of the cord. Tests performed at this time revealed a nonspecific marrow pattern. However, achlorhydria, atrophic gastritis, and the failure to demonstrate the presence of intrinsic factor confirmed the fact that this patient's original macrocytic anemia could be classified as pernicious anemia. Despite frequent injections of refined liver extract he had a red cell count of 3.45 million and a hemoglobin of 9.5 gm./100 ml. at the time of admission. Poor absorption of $B_{12}Co^{60}$ was noted both with and without additional intrinsic factor. This absorption defect could not account for the anemia because the patient had received adequate vitamin B_{12} by way of parenteral liver extract. It is anticipated that further months of thyroid therapy might produce the typical vitamin $B_{12}Co^{60}$ absorption pattern of pernicious anemia. Weekly injections of refined liver, with increasing amounts of thyroid extract over a period of almost three months, had a suboptimal effect on the red cell count (3.92 million/cu. mm.) and hemoglobin (11.5 gm. per cent). Three months later, while on a regimen of 2 cc. of liver extract weekly and 2 gr. of thyroid daily, the red cell count had risen to 4.4 million cells/cu. mm. and the hemoglobin to 13 gm./100 ml. Thus, the anemia was related to the hypothyroid state in the manner of the uncomplicated anemia of hypothyroidism, despite the demonstrated $B_{12}Co^{60}$ test abnormalities.

Extensive studies were not possible in Case III but certain data which we do have are helpful in identifying the type of anemia. Mild normocytic, normochromic anemia with a slightly hypocellular marrow, as was present, is characteristic of the uncomplicated anemia of hypothyroidism. As in the first two cases, decreased absorption of vitamin $B_{12}Co^{60}$ was noted both with and without intrinsic factor. The lack of megaloblastic changes and the failure to show any reticulocytosis or improvement of the anemia on liver injections make it unlikely that this defective absorption was important in the genesis of

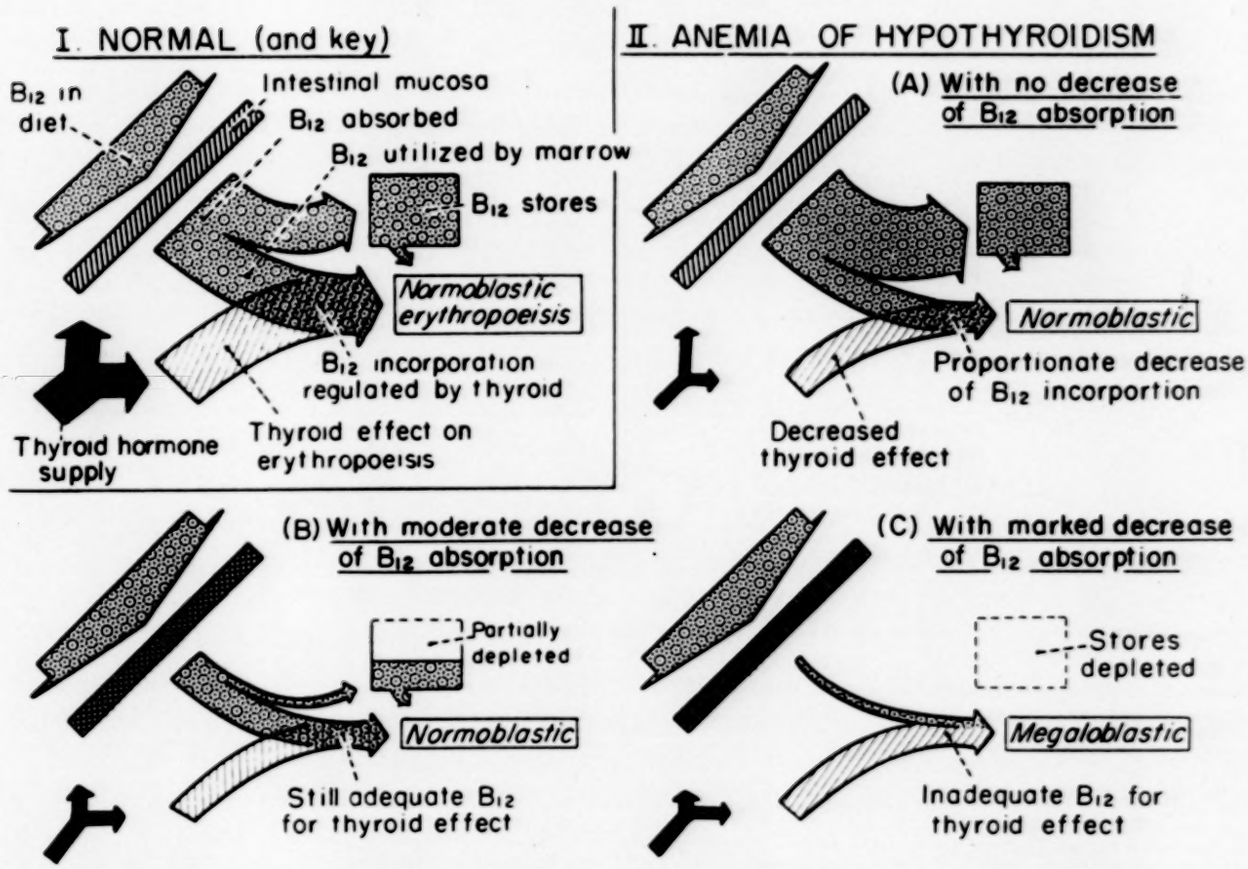

FIG. 3. Diagram of vitamin B_{12} absorption and utilization showing suggested dynamics in the normal subject (I) and in cases of hypothyroidism with no (II A), moderate (II B), and marked (II C) interference with B_{12} absorption secondary to the hypothyroidism. It is noted that depletion of the body vitamin B_{12} stores, which may occur only after very long periods of inadequate absorption, is necessary before megaloblastic anemia develops (II C).

his anemia at this time. However, if the hypothyroidism were allowed to continue, megaloblastic change might well have developed.

Pathophysiology of the Anemias in Hypothyroidism. In Figure 3 we have depicted the probable mechanisms of anemia in hypothyroidism. It is proposed that a deficiency of thyroid hormone has both a direct and an indirect influence on hematopoiesis. The direct influence is much more commonly implicated than the indirect.

Thyroid hormone exerts a direct sustaining action on the marrow which helps to maintain normal cellularity. In hypothyroidism the marrow metabolism is often reduced, with resultant hypocellularity and decreased need for vitamin B_{12}. Whether the lack of thyroid also might partially block the entry or metabolism of vitamin B_{12} in the individual cell (with resultant macrocytosis but not megaloblastosis) is at this time a matter for conjecture. At any rate, this type of decreased erythropoiesis is characteristic of the uncomplicated anemia of hypothyroidism, as noted in Figure 3, II A.

The indirect effect is related to those instances in which a vitamin B_{12} deficiency might occur because of defective intestinal absorption. This would rarely be severe enough to cause a megaloblastic anemia, because the hypocellular marrow would need less vitamin B_{12} than normal. (Figure 3, II B.) In most cases absorption would be improved as a result of thyroid therapy before the body could become sufficiently depleted of B_{12} to result in megaloblastic anemia. Case III would appear to fit such a category. The unusual instance of megaloblastic anemia related to this indirect effect is represented by Case I and by Figure 3, II C.

This indirect effect was found in only three of seven cases studied, and it is assumed that the percentage of all myxedematous patients showing such an abnormality in $B_{12}Co^{60}$ tests would be lower than this. Of cases tested elsewhere,

we know of one showing abnormal tests [*43*], and three giving normal tests [*54*]. Just how hypothyroidism might lead to intestinal malabsorption is another matter for conjecture. Aside from lowering the metabolic activity of all tissues in the body, the myxedematous state causes an increase in the acid mucopolysaccharides of connective tissue ground substance [*59,60*]. It is conceivable that such a deposition might occur in the gastrointestinal tract. The ability to secrete intrinsic factor or to absorb vitamin B_{12} or to do both might be impaired as a result.

SUMMARY AND CONCLUSIONS

Three of seven selected patients with hypothyroidism showed abnormal urinary excretion tests using vitamin $B_{12}Co^{60}$. In all three a low excretion was noted both with and without the addition of intrinsic factor concentrates. Pretreatment with antibiotics did not improve the test in the first two cases. Intrinsic factor was noted to be absent from the gastric juice of these same two. Five attempts were made to demonstrate in their gastric aspirates a substance inhibiting intrinsic factor but in only one such test was there a suggestion of inhibition. Renal function was slightly impaired but did not appear to be a factor in the low excretion of $B_{12}Co^{60}$.

The first patient initially had presented both myxedema and megaloblastic anemia, and with normal gastric acidity. After fifteen (but not nine) months of thyroid therapy he showed slight improvement in control $B_{12}Co^{60}$ tests, a normal test after the addition of intrinsic factor, and some intrinsic factor activity of his gastric juice.

The second case was diagnosed as pernicious anemia many years before hypothyroidism developed. After six months of thyroid therapy both control and intrinsic factor tests remained abnormal.

The third patient with abnormal tests and the four patients showing normal $B_{12}Co^{60}$ excretions were never noted to have megaloblastic anemia.

It is concluded that defective intestinal absorption of vitamin B_{12} sometimes occurs in hypothyroidism, because of a local intestinal block in absorptive ability or depressed secretion of intrinsic factor, or both. These defects, when present, are only very slowly reversible on thyroid therapy. Defective absorption of B_{12} is not the cause of anemia in most cases of hypothyroidism in which thyroid is believed to have a direct effect on erythropoiesis. Decreased demands for vitamin B_{12} by the bone marrow during hypothyroidism may account for the fact that megaloblastic anemia as a complication of myxedema is quite unusual.

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ADDENDUM

Since the preparation of the manuscript, studies have been made upon three additional patients with marked hypothyroidism. One microgram $B_{12}Co^{60}$ was given in each case. One man showed 5.4 per cent twenty-four hour excretion without intrinsic factor; 1.4 per cent with 35 mg. pool 23. Further studies have not yet been made. The two other patients had excretions of 8.1 per cent and 20.0 per cent, respectively.

REFERENCES

1. Best, W. R., Leithold, S. L. and David, D. Urinary excretion of vitamin $B_{12}Co^{60}$ in myxedema. *J. Lab. & Clin. Med.*, 48: 783, 1956.
2. Pitfield, R. L. Myxedema and the nervous system. *Am. J. M. Sc.*, 151: 409, 1916.
3. Mackenzie, S. M. Anemia in hypothyroidism. *J. A. M. A.*, 86: 462, 1926.
4. Stone, C. T. The occurrence of anemia in myxedema. *Ann. Int. Med.*, 2: 215, 1928.
5. Kunde, M. M., Green, M. F. and Burns, G. Blood changes in experimental hypo- and hyperthyroidism (rabbit). *Am. J. Physiol.*, 99: 469, 1932.
6. Bomford, R. Anemia in myxedema, and the role of the thyroid in erythropoiesis. *Quart. J. Med.*, 31: 495, 1938.
7. Eisenberg, S. E. Anemia in hypothyroidism. *Connecticut State M. J.*, 12: 1122, 1948.
8. Chatterjee, S. C. Thyroid and macrocytic hyperchromic anemia. *Calcutta M. J.*, 37: 91, 1940.
9. Sharpe, J. C. and Bisgard, J. D. The relation of the thyroid gland to hematopoiesis. *J. Lab. & Clin. Med.*, 27: 347, 1935.
10. Stern, B. and Altschule, M. D. Hematological studies in hypothyroidism following total thyroidectomy. *J. Clin. Investigation*, 15: 633, 1936.

11. Leonard, M. E., Falconer, E. H. and Ellenhorn, M. J. Anemia of myxedema, with special reference to pernicious anemia. *Ann. Int. Med.*, 34: 1251, 1951.
12. Briggs, J. F. Anemia associated with myxedema. *Journal Lancet*, 75: 541, 1955.
13. Decourt, J., Guillemin, J. and Michard, J. Anemia during myxedema; three cases. *Paris méd.*, 39: 390, 1949.
14. Curschmann, H. The question whether anemia in myxedema is of a pernicious type. *M. Klin.*, 37: 842, 1941.
15. Minot, G. R. Two curable cases of anemia: II. myxedema with anemia. *M. Clin. North America*, 4: 1733, 1921.
16. McCullagh, E. P. and Dunlap, J. H. The blood picture in hyper- and hypothyroidism. In: Crile, G. Diagnosis and Treatment of Diseases of the Thyroid Gland, p. 144. Philadelphia, 1932. W. B. Saunders.
17. Holbøll, S. A. Anemia in myxedema patients. *Acta med. Scandinav.*, 89: 526, 1936.
18. Jones, R. M. Human sternal marrow in hyperthyroid and myxedematous states. *Am. J. M. Sc.*, 200: 211, 1940.
19. Emery, E. S., Jr. The blood in myxedema. *Am. J. M. Sc.*, 165: 577, 1923.
20. Baldridge, C. W. and Greene, J. A. Absence of response of anemia of myxedema to liver extract. *Proc. Soc. Exper. Biol. & Med.*, 31: 1035, 1934.
21. Glass, S. J. Case of myxedema with macrocytic anemia successfully treated with thyroid and testosterone. *J. Clin. Endocrinol.*, 3: 421, 1943.
22. Sturgis, C. C. and Isaacs, R. Treatment of the anemia of myxedema with liver extract. *J. Clin. Investigation*, 8: 663, 1929–1930.
23. Lerman, J. and Means, J. H. Treatment of anemia in myxedema. *Endocrinology*, 16: 533, 1932.
24. Means, J. H., Lerman, J. and Castle, W. B. The coexistence of myxedema and pernicious anemia. *New England J. Med.*, 204: 243, 1931.
25. Barsantini, J. C. and Cervino, J. M. Anemia in myxedema. *Arch. Clin. Inst. Endocrinologia* (*Montevideo*), 2: 119, 1940–1942.
26. Greene, J. A. A comparison of the symptoms, physical and laboratory findings of myxedema and pernicious anemia; with a report of three cases. *Ann. Int. Med.*, 10: 622, 1936.
27. Charaton, F. B. Pernicious anemia associated with thyroid dysfunction: two cases. *St. Thomas's Hosp. Gazette*, 42: 94, 1944.
28. Schneiderbaur, A. The relation between myxedema and pernicious anemia. *M. Klin.*, 37: 659, 1941.
29. Deane, N. and Currence, J. D. Pernicious anemia and hypothyroidism. *New York State J. Med.*, 50: 713, 1950.
30. Bausser, J. and Hellouin de Ménibus, C. Pernicious anemia and myxedema. *Sang.*, 23: 196, 1952.
31. Lisser, H. and Anderson, E. M. Three cases of adult myxedema in women, reported for the purpose of calling attention to their widely different symptomatology and clinical findings. *Endocrinology*, 15: 365, 1931.
32. Vannotti, A. Hypothyroidism, relation to Biermer's anemia. *Schweiz. med. Wchnschr.*, 70: 1106, 1940.
33. Teodori, V. The anemia of myxedema, clinical variants and pathogenetic problems. *Rassegna di neurol. veget.*, 4: 495, 1945.
34. Lerman, J. and Means, J. H. The gastric secretion in exopthalmic goitre and myxedema. *J. Clin. Investigation*, 11: 167, 1932.
35. Mansfeld, S. The thyroid gland and pernicious anemia. *Presse méd.*, 46: 993, 1938.
36. Okuda, K., Steelman, S. and Chow, B. F. Absorption of B_{12} in hyperthyroid and hypothyroid rats. *Federation Proc.*, 15: 567, 1956.
37. Schilling, R. F. Test for intrinsic factor activity. *J. Lab. & Clin. Med.*, 42: 946, 1953.
38. Best, W. R., White, W. F., Robbins, K. C., Landmann, W. A. and Steelman, S. L. Studies on urinary excretion of vitamin $B_{12}Co^{60}$ in pernicious anemia for determining effective dosage of intrinsic factor concentrates. *Blood*, 11: 338, 1956.
39. Best, W. R. Diagnostic tests using vitamin B_{12} labeled with cobalt^{60}. In: Fields, T. and Seed, L. Clinical Use of Radioisotopes, p. 62. Chicago, 1957. Year Book Publishers.
40. Ellenbogen, L., Williams, W. L., Rabiner, S. F. and Lichtman, H. C. An improved urinary excretion test as an assay for intrinsic factor. *Proc. Soc. Exper. Biol. & Med.*, 89: 357, 1955.
41. Fields, T. and Seed, L. Clinical Use of Radioisotopes. Chicago, 1957. Year Book Publishers.
42. Bishopric, G. A., Garrett, N. J. and Nicholson, W. M. Clinical value of the TSH test in the diagnosis of thyroid diseases. *Am. J. Med.*, 18: 15, 1955.
43. Schwartz, S. O. and Maslow, W. Personal communication.
44. Wolff, R. and Weiss, J. The possible role of gastric juice in protecting B_{12} from chemical destruction. *Rev. d'hémat.*, 10: 202, 1955.
45. Davis, L. J. and Brown, A. The Megaloblastic Anemias, p. 36. Springfield, 1953. Charles C Thomas.
46. Girdwood, R. H. The possible effects of gastrointestinal bacteria on the absorption of vitamin B_{12}. *Rev. d'hémat.*, 2: 187, 1955.
47. Halsted, J. A., Lewis, P. M. and Gasster, M. Absorption of radioactive B_{12} in the syndrome of megaloblastic anemia associated with intestinal stricture or anastomosis. *Am. J. Med.*, 20: 42, 1956.
48. Rath, C. E., McCurdy, P. R. and Duffy, J. B. Effect of renal disease on the Schilling test. *New England J. Med.*, 256: 111, 1957.
49. Cohn, C. Endocrine influences on renal function. Progress in Clinical Endocrinology. New York, 1950. Grune & Stratton.
50. Yount, E. and Little, J. M. Renal clearance in patients with myxedema. *J. Clin. Endocrinol.*, 15: 343, 1955.
51. Heinle, R. W., Welch, A. D., Scharf, V., Meacham, G. C. and Prusoff, W. H. Studies of excretion (and absorption) of Co^{60}-labeled vitamin B_{12} in pernicious anemia. *Tr. A. Am. Physicians*, 55: 214, 1952.
52. Doscherholmen, A. and Hagen, P. S. Radioactive vitamin B_{12} absorption studies; results of direct measurement of radioactivity in the blood. *Blood*, 12: 336, 1957.
53. Glass, G. B. J., Boyd, L. J., Gellin, G. A. and Stephanson, L. Uptake of radioactive vitamin B_{12}

by the liver in humans: test for measurement of intestinal absorption of vitamin B_{12} and intrinsic factor activity. *Arch. Biochem.*, 51: 251, 1954.

54. Miller, A., Corbus, H. F. and Sullivan, J. F. A modified urinary excretion test for measuring oral cobalt^{60} labeled vitamin B_{12} absorption and its application in certain disease states. *Blood*, 12: 347, 1957.
55. Miller, O. N. and Unglaub, W. G. A study of etiology of macrocytic anemia. *J. Clin. Investigation*, 36: 916, 1957.
56. Stevenson, T. D., Little, J. A. and Langley, L. Pernicious anemia in childhood. *New England J. Med.*, 255: 1219, 1956.
57. Wintrobe, M. M. Clinical Hematology, 3rd ed., p. 449. Philadelphia, 1956. Lea & Febiger.
58. Rubin, C. E. Observations on the pathogenesis of juvenile pernicious anemia. *J. Clin. Investigation*, 36: 925, 1957.
59. Watson, E. M. and Pearce, R. H. M. S. The mucopolysaccharide content of the skin in localized (pretibial) myxedema. *Am. J. Clin. Path.*, 19: 442, 1949.
60. Wiener, R., Iannaccone, A., Eisenberg, J., Griboff, S. I., Ludwig, A. W. and Soffer, L. J. Influence of hormone therapy on body fluids, electrolyte balance and mucopolysaccharides in myxedema. *J. Clin. Endocrinol.*, 15: 131, 1955.

Fenestration of the Semilunar Cusps, and "Functional" Aortic and Pulmonic Valve Insufficiency*

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IN the remarkable treatise published more than 100 years ago, James Hope [*1*] wrote "Another affection of the valves, whether auricular or semilunar occasioning regurgitation, is atrophy. By this, I have seen the membranous expansion of the mitral valve reduced to a mere reticulated web and the aortic valves perforated in five or six places. The affection commonly occurs in connection with general atrophy and anemia."

Rokitansky [*2*] gave an accurate account of valvular fenestration, a condition which he regarded as a form of atrophy. Very little can be added to his original description which follows: "These perforations are almost always situated near the free margin of the valves, and more especially near their insertion, where they originate, increasing in numbers as they spread towards the nodules of the valve. They are at first about the size of a scarcely appreciable pin-hole, or of a poppy-seed, but after gradually enlarging by the confluence of several into one, they finally attain the size of a grain of millet, or of a hempseed, or even of a pea. When several are present together they impart a reticular broken appearance to the valve. The perforations are moreover surrounded by a smooth margin, and are never round, but oval, elliptical, or fissure-like and their long axis is at right angles to the free margin of the valve. They are also generally bounded by the fibrous bundles of the valves so that the atrophy, at least at first, attacks only the thinnest portions. Besides considerable and appreciable attenuation of the valve, and in some cases even perforation, we occasionally find some portions, as for instance the free margin, the nodule, and the fibrous bundles passing from it thickened and hypertrophied."

This valvular lesion has engaged very little attention on the part of clinicians and pathologists. Many pathologists do not record its existence in routine autopsy protocols, partly because of failure to recognize it, and also because of the belief that it is a prevalent finding possessing no clinical significance.

Foxe [*3*] reported a statistical study in 300 successively observed hearts at Bellevue Hospital. Fenestration was present in 82 per cent involving the aortic and pulmonic leaflets almost equally. Isolated cases have been reported mainly in connection with spontaneous rupture of the leaflets, or with the appearance of a diastolic murmur [*4–6*].

Our interest in this disorder was stimulated when we observed two patients, who had systemic hypertension, well defined aortic diastolic murmurs, and fenestrated aortic cusps. In one patient, the existence of the valvular defect was suspected before death. The present report is concerned with these subjects and with observations on the appearance of the semilunar valves in 342 autopsies performed at the University and at the Veterans Hospitals.

CASE REPORTS

CASE. 1 A fifty-seven year old Negro man noted dyspnea on effort which began two months prior to admission and progressed rapidly to nocturnal dyspnea, orthopnea, cough, and blood tinged sputum. He denied having had a penile lesion, but thought that he received some injections for "bad blood" five or eight years earlier. For several years a physician

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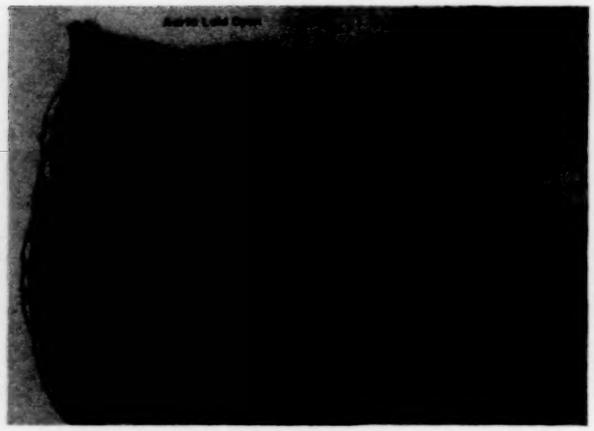

FIG. 1A. Case I, drawing showing defects in two aortic leaflets and adjoining commissure.

FIG. 1B. Case II, fenestration in all three aortic leaflets.

placed him on a low salt diet because of "high blood pressure." For one year he complained of aching and slight swelling in the left shoulder, aggravated by movement of the shoulder. Some fluid was withdrawn from the shoulder region. A diagnosis of subdeltoid bursitis and hypertension was made. The blood pressure recorded one year previously was 218/112 mm. Hg. The heart was considered to be enlarged and systolic, as well as diastolic murmurs recorded in the aortic area. Cardiac enlargement and proteinurea 2 plus was noted. Blood serologic tests for syphilis were negative.

On the last admission, he appeared markedly dyspneic. The heart was enlarged, the apical impulse being felt in the 6th intercostal space in the anterior axillary line. There was a loud grade 3 systolic, as well as grade 3 diastolic, murmur heard best at the aortic area and transmitted over the entire precordium. A definite Austin Flint murmur was not detected. The pulses were bounding. Blood pressure was 200/64 mm. Hg. There was pulmonary congestion, venous distention, hepatic enlargement and slight pretibial edema. The fundus showed moderate arterial narrowing and arteriovenous nicking. The electrocardiogram was compatible with the findings displayed in left ventricular hypertrophy and strain. Proteinuria and nitrogen retention were persistently present. The blood urea nitrogen was 92 mg. per cent. Death occurred in a state of uremia and congestive heart failure. Although serologic tests for syphilis were consistently negative, the most probable diagnosis was considered to be syphilitic aortic insufficiency, complicated by renal hypertension.

The heart weighed 560 gm., the predominant hypertrophy being in the left ventricle. The ascending aorta was slightly dilated and measured 8 cm. in circumference. There was marked atherosclerosis of the arch and descending aorta, but no gross or microscopic evidence of syphilis was noted. The coronary ostia and vessels were patent. The left circumflex branch was enlarged, and tortuous, and aneurysmal, measuring 2.5 cm. in circumference.

The left anterior and posterior cusps showed large fenestrations, measuring 1 by 1.5 cm. in each cusp. (Fig. 1A.) The defect involved part of the commissure so that complete closure was impaired. The edges were smooth and not jagged, and there was no evidence of inflammation or tear. Hematoxylin and eosin stained section of aortic valve through the margins of the defect showed the connective tissue to have a diffuse hyaline, structureless, pale eosinophilic matrix, save for small focal pale hematoxylin staining areas resembling myxomatous change in the central portion. For the most part, the connective tissue fibrils were not discernible, but there were scattered round to oval tissue nuclei. No inflammatory reaction was noted. The kidneys were reduced in size and showed changes of chronic pyelonephritis.

Summary. A Negro man (age 57) had pyelonephritis and known hypertension for at least one year, terminating in rapidly progressing heart failure and renal failure. Auscultatory and peripheral signs of free aortic insufficiency were present, and undoubtedly contributed to the cardiac strain. The diagnosis of syphilitic aortic insufficiency figured most prominently in the differential diagnosis. The lesion proved to be noninflammatory fenestration of aortic valve cusps.

CASE II. A fifty-seven year old Negro laborer was first observed in September 1949 complaining of nocturia and periods of mental confusion. In 1918, he had an attack of gonorrhea. A small penile lesion was present at the same time, but he denied having had a diagnosis of or treatment for syphilis. In 1937, he complained of backache. Records from another hospital of the examination at that time showed moderate adiposity, normal sized heart, no murmurs, blood pressure 130/86 mm. Hg, normal urine with maximum specific gravity of 1.026. Serologic tests for syphilis were negative. Twelve years later, the perti-

nent findings on physical examination were moderate obesity, moderate cardiac enlargement, retinal arteriosclerosis, prostatic hypertrophy and minimal pretibial edema. The second aortic sound was loud and ringing. There was a grade 2 systolic murmur and a grade 2 blowing decrescendo diastolic murmur in the aortic area transmitted down the sternum. The blood pressure ranged between 254/140 and 170/108 mm. Hg. It was noted that the intensity of the murmur varied directly with the level of the diastolic pressure. After inhaling amyl nitrate, the blood pressure declined from a level of 190/110 to 140/80 mm. Hg. The murmur could not be heard at all at the lower level for a period of about ninety seconds, and reappeared as the pressure returned to the original point. On several occasions, it was observed to be present at pressure levels, as low as 160/96 mm. Hg. The electrocardiogram showed changes compatible with left ventricular hypertrophy. Renal function was impaired, the maximum concentration being 1.017 and maximum urea clearance 23 per cent of normal. Serologic tests for syphilis were negative in blood and spinal fluid. The clinical diagnosis was essential hypertension, with functional aortic insufficiency and arteriolar nephrosclerosis. The existence of fenestration in the aortic cusp was postulated. Since syphilitic aortitis could not be absolutely excluded the patient was treated with 6,000,000 units of penicillin.

In February 1953, he was again observed during an episode of mild congestive heart failure. At this time, the blood pressure was 240/130 mm. Hg, and a grade 2 aortic diastolic murmur was again recorded. There was evidence of further decline in renal function. He improved on medical management and got along fairly well until July 1954, when hemiparesis developed followed by lobar pneumonia. On the final admission, the blood pressure varied between 200/120 and 180/100 mm. Hg. The systolic, as well as diastolic, murmurs were again noted. The pneumonic process responded to antibiotic management. Urea nitrogen levels in the blood varied between 20 and 40 mg. per cent. The neurologic symptoms progressed, convulsions and coma developed and the patient died.

Postmortem examination revealed cerebral arteriosclerosis with cystic encephalomalia of the pons, medulla and ventrolateral mid-brain, arteriolonephrosclerosis and bilateral bronchopneumonia.

The heart weighed 450 gm., the thickening being predominantly in the left ventricle. The aortic and pulmonic rings each measured 8 cm. in circumference. All three aortic cusps showed fenestrations at the free overlapping margins adjacent to the commissures. (Fig. 1B.) The aorta was slightly dilated and there were small atheromatous plaques in the ascending portion. There was slight coronary atherosclerosis but no myocardial infarction. There was no gross or microscopic evidence of syphilis in the valves or aorta.

Summary. This patient had essential hypertension for at least five years. Throughout this period a murmur of aortic insufficiency was recorded. The intensity of the murmur varied with levels of diastolic pressure and could be made temporarily inaudible by inhaling amyl nitrite. The existence of significant aortic valve fenestration was suspected before death. The aortic valves showed fenestration at apposing margins in all three cusps.

AUTOPSY MATERIAL AND METHOD

The clinical and autopsy records in two institutions were reviewed and the heart valves carefully examined with particular reference to fenestration. Subjects with congenital cardiovascular lesions, or with organic disease, or defects of the semilunar valves other than fenestration were excluded. At the Veterans Hospital, there were 159 male patients, ranging from twenty-two to eighty years of age, all of whom had died within the previous year. At the University Hospital, there were 106 men and seventy-seven women whose hearts were available for examination. They ranged in age from one day to ninety-five years. There were 208 white and 134 Negro subjects.

The measurements of the heart weights and the aortic and pulmonic outlets were taken as recorded in the fresh specimen by many different prosectors. Similarly, the presence or absence of diastolic murmurs and the level of blood pressure is based on the recordings of many observers.

The degree of fenestration was estimated as the sum of the longest diameter of the defects. The lace-like reticulated lesion was measured in the overall length as though it were a single defect. Since the fenestrations varied in appearance from round or oval holes to mere fissures consisting of multiple small adjoining openings which could be altered by extending or stretching the free edge, the measurement recorded provides only a rough indication of the actual area of the defect.

Studies were made of longitudinal and transverse sections of pulmonic and aortic valve cusps of fifty-one men ranging in age from twenty-two to seventy-two years. Hematoxylin and eosin, alcian blue-PAS and orcein–van Gieson stained sections were studied in all of the cases; Masson's trichrome stained sections in ten cases and Sudan IV stained gross tissues in ten cases.

RESULTS

In the 342 hearts examined, 247 or 72 per cent showed some degree of fenestration. (Table I.)

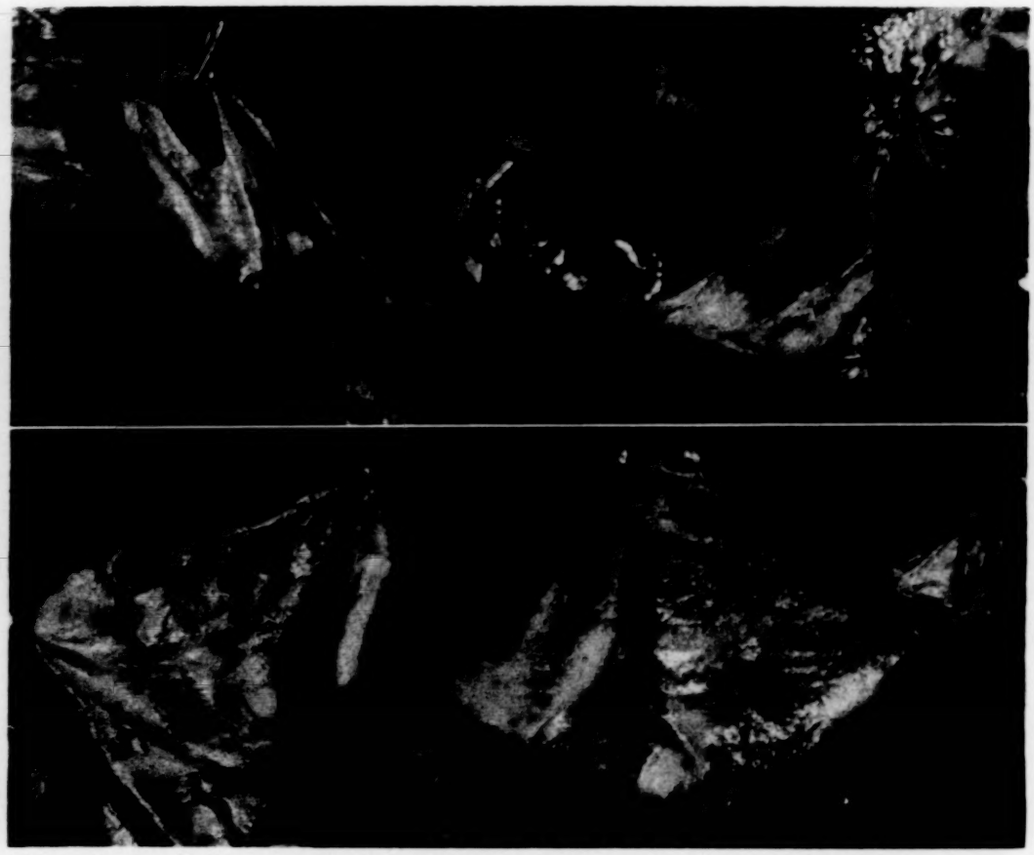

FIG. 2. A sixty-eight year old man with cor pulmonale, chronic fibrosis and emphysema, and bronchogenic carcinoma. Grade 2 diastolic murmur at pulmonic area. Heart weight was 410 gm.; blood pressure, 118/60 mm. Hg. Top, aortic valve, circumference 8.5 cm. Fenestration in two cusps. Bottom, pulmonic valve, circumference 9.5 cm. Fenestration in two cusps.

The aortic valve was affected slightly more frequently than the pulmonic, but each valve was involved in more than half the subjects. The two valves were affected together about twice as often as they were independently. Figure 2 illustrates an instance of moderately severe fenestration of both valves in the same individual.

TABLE I
INCIDENCE OF FENESTRATION IN AORTIC AND PULMONIC VALVES

Data	No. of Patients	Per cent
Fenestration absent	95	28
Fenestration present	247	72
Aortic valve only	70	20
Pulmonic valve only	57	17
Aortic and pulmonic valves	120	35
Total aortic valve fenestrations	190	55
Total pulmonic valve fenestrations	177	52
Total patients	342	100

In more than one-third of the instances of fenestration, only a single cusp was affected. The number of cusps involved declined progressively (Table II) to 4 per cent or eleven patients in whom all six leaflets were fenestrated. The distribution of the lesions in the individual cusps are illustrated graphically in Figure 3.

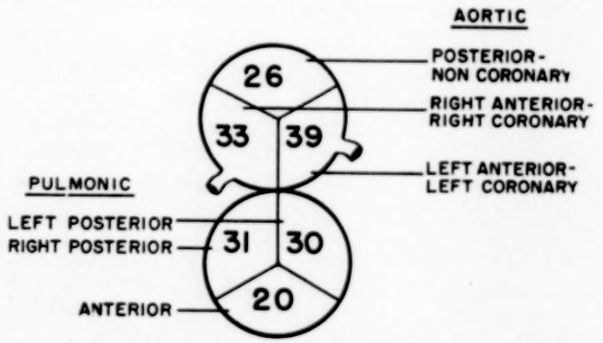

FIG. 3. Distribution of fenestration in individual semilunar cusps. The numbers denote per cent involvement of the respective leaflet in all 342 hearts.

The area of the individual defects varied widely from pinhead size at one extreme to one-fifth of the surface of the cusps at the other. The sum of the lengths in the valvular fenestrations varied from 1 to 60 mm., the average being slightly bigger in the aortic (13.3) than in the pulmonic valve (10.8).

TABLE II
INCIDENCE OF FENESTRATED CUSPS

No. of Cusps Affected	Valves Affected No. of Cases			
	Aortic	Pulmonic	Aortic and Pulmonic	Total No. of Patients Affected (%)
1	47	44	...	91 (37)
2	17	11	24	52 (21)
3	6	2	34	42 (17)
4	..	..	29	29 (12)
5	..	..	22	22 (9)
6	..	..	11	11 (4)
Total No. of Cases	70	57	120	247 (100)

The incidence and severity of fenestration was about the same for Negro and white races, but was considerably higher in men than women. (Table III.) The male subjects were on the average five years older than the women. This age difference does not account for the striking variation in valvular defects. In general, the older age groups showed a higher incidence of fenestration. (Table IV.) This was not consistent. The X^2 value for semilunar valve lesions is 12.8 with a P value 0.1. It is noteworthy that even in the first decade of life, thirteen out of twenty hearts, or 65 per cent, had demonstrable fenestrations in the semilunar valves. In children, these openings were tiny consisting sometimes of only one or two isolated pinhead-sized holes. In older subjects, they tended to enlarge both in number and area. As measured by the longest diameter, there appeared to be an increase in the average size of the opening with advancing age.

TABLE III
INCIDENCE OF FENESTRATION IN AORTIC AND PULMONIC VALVES

No. of Patients Examined	Men	Women
	265	77
No. (%) with fenestrated aortic and/or pulmonic valves	203 (76)	44 (57)
No. (%) with fenestrated aortic valves	162 (61)	28 (36)
Degree of aortic fenestration, average length (mm.)	14	9
No. (%) with fenestrated pulmonic cusps	147 (55)	30 (39)
Degree of pulmonic fenestration, average length (mm.)	12	8
Average age in years	50	45

TABLE IV
FENESTRATION IN RELATION TO AGE

Age (yr.)	No. of Cases	Aortic Valve Fenestration		Pulmonic Valve Fenestration	
		Frequency (%)	Length (mm.)	Frequency (%)	Length (mm.)
To 10	20	45	4	30	5
10–20	9	55	9	44	8
20–30	19	32	11	42	7
30–40	34	59	12	56	10
40–50	54	61	13	63	12
50–60	62	60	13	55	11
60–70	99	59	16	55	12
70–over	45	48	14	40	13

The left anterior aortic leaflet showed the highest incidence and the anterior pulmonic cusp the lowest incidence of fenestration. (Fig. 3.) The right anterior aortic and right posterior pulmonic leaflets arise from a common primitive fold [7]. The left anterior aortic and left posterior

TABLE V
DISTRIBUTION OF LEAFLET FENESTRATION IN 120 CASES OF COMBINED AORTIC AND PULMONIC INVOLVEMENT

Pulmonic Leaflet Involved	Aortic Leaflet Involved—No. of Patients		
	Right Anterior	Left Anterior	Posterior
Right posterior	44	62	52
Left posterior	46	60	50
Anterior	29	40	40

pulmonic cusps are likewise paired embryologically. The influence of a common anlage is reflected in Table v which shows the distribution of fenestrated leaflets in the 120 hearts in which there was simultaneous involvement of both semilunar valves. It shows that the embryologically paired cusps are not affected to any greater extent than are the opposite unpaired leaflets in the same heart.

TABLE VI
FENESTRATION IN RELATION TO HEART WEIGHT

Weight (gm.)	No. of Cases	Aortic Valve Fenestration		Pulmonic Valve Fenestration	
		Frequency (%)	Length (mm.)	Frequency (%)	Length (mm.)
To–300	79	49	11	42	12
300–400	101	52	15	54	12
400–500	83	61	14	55	10
500–600	37	60	11	53	14
600–700	22	64	15	50	8
700–over	13	69	15	61	12

Table VI shows the relationship of heart weight to fenestration. With augmenting heart weights, there is an increasing frequency but not severity of fenestration in the aortic leaflets. In the pulmonic leaflet this trend is not apparent except with the heaviest hearts exceeding 700 gm.

TABLE VII
FENESTRATION IN RELATION TO SYSTEMIC AND PULMONARY HYPERTENSION

Data	Systemic Circulation		Pulmonary Hypertension *
	Normotensive *	Hypertension *	
No. of patients	187	68	23
(%) fenestration aortic valve	51	65	61
(%) fenestration pulmonic valve	52	57	68
Length defect (mm.) aortic valve	14	14	18
Length defect (mm.) pulmonic valve	12	11	16

* For criteria of selection, see text.

A similar relationship was observed when the data were charted with respect to systolic and diastolic blood pressure levels. A comparison of the findings in subjects with pulmonary and systemic hypertension is summarized in Table VII. The criteria for systemic hypertension was a diastolic pressure of at least 100 mm. Hg and a systolic of 150 or higher. The criterion for the normotensive group was a diastolic pressure below 90 mm. Hg. The category designated "pulmonary hypertension" consisted of individuals who had pronounced disproportionate right ventricular hypertrophy with a pulmonary or mitral valve lesion sufficient to result in chronic right ventricular strain. This section consisted of seventeen patients with chronic obstructive emphysema, two with diffuse granulomatous disease of the lung, two with advanced silicotuberculosis, and fibrosis, and two persons with severe mitral stenosis. It may be seen that in the hypertensive subjects, whether systemic or pulmonary, there is an increased incidence of fenestration which affects both semilunar valves but is greater in the leaflets that are under stress. The average size of the valvular defect is unchanged in the systemic hypertensive subject but increased in the subjects with lesions leading to pulmonary hypertension.

TABLE VII
FENESTRATION IN RELATION TO CIRCUMFERENCE OF AORTA AND PULMONARY ARTERY

Circumference (cm.)	Aortic Valve Fenestration		Pulmonic Valve Fenestration	
	Frequency (%)	Average Length (mm.)	Frequency (%)	Average Length (mm.)
Aortic outlet:				
6–7	44	11	41	8
7–8	57	12	53	11
8–9	63	16	55	13
9 and over	72	20	77	16
Pulmonic outlet:				
6–7	41	11	43	9
7–8	51	13	49	12
8–9	63	15	52	12
9 and over	71	18	68	13

In Table VIII, the values are charted in relation to circumference of the aortic and pulmonary artery outlets. It is evident that with

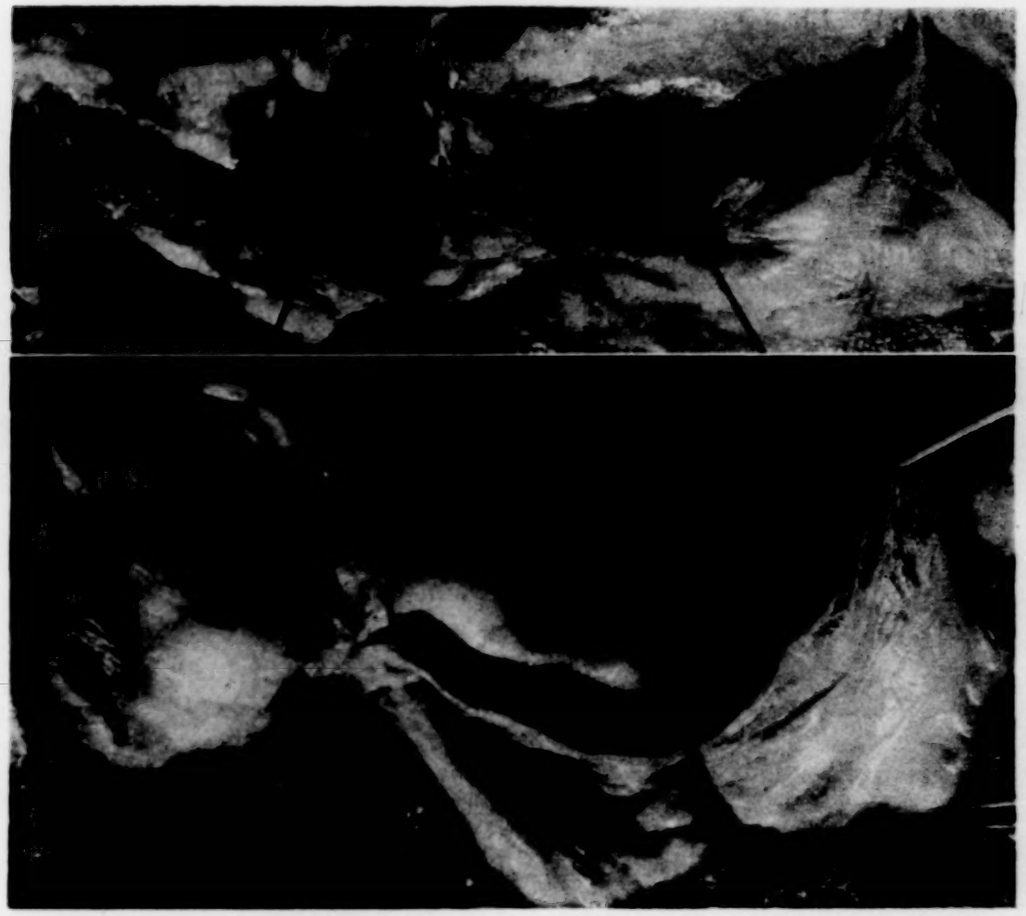

FIG. 4. Top, a sixty-six year old man with carcinoma of the bladder, hypertension, myocardial infarction and small ventricular aneurysm. Circumference of the aorta, 9.0 cm. Diastolic murmur over precordium. Bottom, a thirty-seven year old man with rheumatic heart disease, mitral stenosis and Graham Steell murmur. Heart weight, 830 gm.; circumference of the pulmonary artery, 9.5 cm.

widening of either ring, there is a definite increase in severity and in frequency of fenestration in both of the semilunar valves.

Microscopic study revealed no constant change, save for a gradual decrease in cellularity of the aortic valve leaflet with age. This change was much less apparent in the pulmonic valve. Almost all sections showed some degree of hyalinization of the dense collagenous connective tissue and obscuring of the fibrils. In addition, the loose connective tissue showed varying degrees of vacuolization. The formalin fixed tissues from the ten cases stained with Sudan IV all showed linear mottled positive sudanophilic material, paralleling the bundles of connective tissue, presumably occupying the vacuolated spaces in the loose connective tissue of the valve leaflets. The ground substance showed varying degrees of intensity of staining with alcian blue. In general, the loose connective tissue exhibited a greater amount of alcian blue positive material than the remainder of the cusp. Several of the pulmonic and aortic leaflets showed myxomatous change of varying degrees. No definite correlation could be made between any of the changes noted above and the presence of fenestration. The normal valves demonstrated the same range of change as did the fenestrated valves.

Diastolic murmurs at the base of the heart were recorded in only four instances, three of which were pulmonic, and one probably aortic in origin. Two of the pulmonic murmurs were observed in subjects with mitral stenosis (Graham-Steell murmur), one of whom showed fenestrated cusps (Fig. 4, bottom), and the other intact pulmonic valves. The third instance of pulmonary diastolic murmur was noted in a patient with cor pulmonale due to chronic pulmonary fibrosis and emphysema. Fenestration of the pulmonic and aortic leaflets was

present (Fig. 2, bottom). The aortic diastolic murmur was observed in a sixty-six year old man who died of carcinoma of the bladder with extensive metastasis. Hypertension and myocardial infarction were recorded sixteen years earlier. Three months prior to death, he had a grade 2 blowing diastolic murmur over the mid-sternum and left precordium. Blood pressure was 200/106 mm. Hg. At necropsy, the heart weighed 550 gm. and showed moderate fenestration of all three aortic leaflets. (Fig. 4, top.) The aorta showed atherosclerosis and calcification in the ascending portion. In the left ventricle, there was an area of old infarction with aneurysmal dilatation measuring 1.5 cm. in diameter. In this individual it is possible that the small ventricular aneurysm, and not the fenestration, was responsible for the murmur.

COMMENTS

The observations in this autopsy series are in close agreement with those recorded by Foxe. Fenestrated semilunar valves are present in more than 70 per cent of the hearts examined. It affects Negro and white races equally, men more than women. The pulmonic valve is involved almost as often as the aortic, but with lessened severity as measured by the length of the gaps rather than by the number of holes. Why some leaflets are affected more than others is not apparent. It is noteworthy that the two cusps that have fewest defects (Fig. 2) are those that develop from independent primitive buds that are farthest removed from the truncus ridge. They are subject to less strain than the lateral valves which are attached to the aortopulmonary septum and which are exposed to the torsion imparted to the common wall by two independent streams. The other four cusps develop from two lateral buds, each fold contributing to both aortic and pulmonic valves. This circumstance might be expected to predispose to parallel involvement of embryologically paired cusps if developmental defects were a major etiologic factor. Actually no such correlation was observed in charting the distribution of fenestrated leaflets in those hearts which showed lesions in both semilunar cusps. Foxe found fenestration to be present in fetal hearts and it was not correlated with patency of the foramen ovale. Nevertheless, a congenital tissue defect as an etiologic factor cannot be excluded on this ground.

The most impressive correlation in this series of hearts is with the circumference of the aortic and pulmonic rings on the one hand and degree of fenestration in both semilunar valves on the other. This observation suggests that there is a common factor which may underlie both the valvular and large vessel lesions. Increased vascular tension may play some part as evidenced by the slightly greater involvement of the aortic leaflets in systemic hypertension and of pulmonic cusps in pulmonary hypertension. But it does not account for concommitant increment in defect in the valve not under stress.

The exact cause of the lesion is not clear. There is no histologic evidence of inflammation. Congenital anomaly cannot be entirely excluded but does not seem likely. The incidence of fenestrated valves in this series, from which the usual varieties of congenital heart disease was excluded, is slightly higher than in a group of 148 cases of congenital cardiac abnormalities reported by Simpson [*8*].

We believe that it is a form of atrophy which may begin in early childhood, or even in the fetus, and that aging, dilatation of the ring and increased intravascular pressure are contributing and modifying factors.

Functional diastolic murmurs occurring at the base of the heart have been recognized for many years. They have been associated with systemic and pulmonary hypertension, high fever, severe anemia, thyrotoxicosis and other hyperkinetic circulatory states. Graham Steell [*9*], in the classical account of the murmur which bears his name, refers to a description of a similar murmur in the aortic valve associated with aortic dilatation. Garvin [*10*] reported fourteen cases of aortic diastolic murmurs in a survey of 200 consecutive cases of hypertensive heart disease. Paullin and his co-workers [*11*] found a diastolic aortic murmur in 2.4 per cent of hypertensives. The Graham Steell murmur has been regarded as the classical murmur of functional insufficiency of the pulmonary valve.

Most instances of fenestration of the semilunar cusps are clinically unimportant. Occasionally, they give rise to diastolic murmurs, which are confused with the murmurs of organic valve disease. When hypertension, aortic dilatation and chest pain are present, as they frequently are, the picture may mimic dissecting aneurysm.

Dynamically important aortic regurgitation may rarely develop as a consequence of rupture of the connecting tissue bands and enlargement of the defect. The patient in Case 1 in this report

had free aortic regurgitation without gross evidence of a torn leaflet. In this case, there was a history of antecedent syphilis. A diagnosis of luetic heart disease was erroneously made despite the negative conventional serologic tests. The Treponema pallidum immobilization tests are helpful in differential diagnosis in such situations, since they are generally reactive in otherwise seronegative patients with syphilitic heart disease [*12*]. Since the gaps in the leaflets are present so frequently, why don't we hear the murmurs more often? We believe there are several explanations. The fenestrations which are located mainly at the commissures and at the thin pliable lunulae at the margins are closed off in diastole by apposition of adjacent leaflets. Under ordinary circumstances, regurgitation would occur only if the defect involved the bodies or the non-apposing portion of the cusp. Under conditions of hypertension or dilatation of the ring, failure of the closing surfaces to meet may give rise to backflow with lesser degrees of fenestration. Such could well be the situation in Case II and in three of the four instances of diastolic murmurs noted in the statistical study. It is also highly probable that minor degrees of valvular regurgitation are below the auscultatory threshold. We have observed in the dog aortic valvular insufficiency, the presence of which could be proved unequivocally by a salt injection method, but which produced no audible murmurs even when the stethoscope was applied directly to the surface of the heart.

The coexistence of the two conditions does not necessarily mean that the valvular fenestration is responsible for the diastolic murmur. When hypertension is also present, one can never be certain whether the blood flows back through the holes in the leaflets or the gap at the closing margin of the cusps. Functional aortic and pulmonic murmurs may occur in the absence of fenestration. This was in fact found in one of the two subjects with mitral stenosis and Graham Steell murmur. In the other five cases, the diastolic murmur was accompanied by moderate to severe degrees of fenestration. In those instances, the term "functional" murmur cannot be applied in the strict sense. Here there is an actual anatomic defect in the cusps potentially capable of limiting the holding ability of the valve. Insufficiency is realized under conditions of increased intravascular pressure and stretching of the ring

Most patients with hypertension do not have aortic diastolic murmurs. If we assume that the appearance of the murmur is conditioned by the presence of large fenestrations, we may readily explain why it is found in some and not in others with equally severe, or even higher levels of blood pressure. The same reasoning applies to the pulmonic diastolic murmur.

The "functional" diastolic murmur is encountered more often in connection with pulmonary than with systemic hypertension even though the degree of fenestration is on the average slightly less in the pulmonic than the aortic valves. The explanation may reside in the differences in relative increments in the pressures against which the valves must hold in pathologic states. While the absolute aortic diastolic pressures exceed the pulmonic, the rise in pressure in the aorta seldom exceeds two, and is never more than three times the normal level, whereas in the pulmonary artery, diastolic pressure may rise six fold or more.

The valvular lesion is commonly overlooked in routine autopsies, or else regarded as a normal state not worthy of mention. Its existence was reported in the routine autopsy protocols in less than 1 per cent of the hearts in which it was actually present. More careful scrutiny of the valve surface and recording of the pathologic and clinical observations will help to determine the respective roles of structural and functional factors in the mechanism of the valvular insufficiency.

SUMMARY

1. Two cases, and a possible third case, are described in which fenestrated aortic leaflets in hypertensive patients were associated with diastolic murmurs. Well defined fenestration of the pulmonary cusps was noted in two out of three patients with hypertension of the pulmonary circuit and a Graham Steell murmur.

2. Fenestrated semilunar valves were found in 72 per cent of 342 hearts examined. The findings were reviewed with respect to age, sex, leaflets involved, and relation to cardiac hypertrophy, intravascular pressure, and dilatation of the outlet, and the presence of diastolic murmurs.

3. It is suggested that the functional diastolic murmurs of the aortic and pulmonic valves may in some instances be due to the presence of anatomically defective valve leaflets which, under conditions which lead to dilatation of the ring, permit sufficient backflow to result in the sound which is audible over the chest wall.

REFERENCES

1. Hope, J. A Treatise on the Diseases of the Heart and Great Vessels. p. 346. Philadelphia, 1842. Lea and Blanchard.
2. Rokitansky, C. Handbuch der pathologische Anatomie. Translated by George E. Day. Vol. 4, p. 228. London, 1852. Sydenham Society.
3. Foxe, A. N. Fenestrations of the semilunar valves. *Am. J. Path.*, 5: 179, 1929.
4. Castleman, B. and Towne, V. W. Case record of Massachusetts General Hospital. Marked fenestration of aortic cusps leading to aortic regurgitation. *New England J. Med.*, 245: 941, 1951.
5. Mathews, R. J. and Darvill, F. T., Jr. Fenestrations of the aortic valve cusps as a cause of aortic insufficiency and spontaneous aortic valve cusp rupture. *Ann. Int. Med.*, 44: 993, 1956.
6. Prodfit, W. L. and McCormack, L. J. Rupture of aortic valve with fenestration. *Circulation*, 13: 750, 1956.
7. Arey, L. B. Dèvelopmental Anatomy. Philadelphia, 1941. W. B. Saunders Co.
8. Simpson, F. O. Congenital abnormalities of the heart in the insane. *J. Anat. & Physiol.*, 32: 679, 1897–1898.
9. Graham Steell. The murmurs of high pressure in the pulmonary artery. *Medical Chronicle* (Manchester), 9: 182–188, 1888.
10. Garvin, C. F. Functional aortic insufficiency. *Ann. Int. Med.*, 13: 1799, 1940.
11. Paullin, J. E., Bowcock, H. M. and Wood, R. H. Complications of hypertension. *Am. Heart J.*, 2: 613, 1927.
12. Friedman, B. and Olansky, S. Diagnosis of syphilitic heart disease with special reference to T. P. I. tests. *Am. Heart J.*, 50: 323, 1955.

Congenital Absence of a Main Branch of the Pulmonary Artery*

Report of Three New Cases Associated Respectively with Bronchiectasis, Atrial Septal Defect and Eisenmenger's Complex

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ANGIOCARDIOGRAPHY has made possible the diagnosis of congenital absence of a main branch of the pulmonary artery during life whereas previously the diagnosis was made only at necropsy or operation. The first angiocardiographic study of a patient with congenital absence of the right pulmonary artery was carried out in 1948, and, together with another study of a patient with an absent left pulmonary artery, was reported in 1953 [*1*]. However, the first published example of the anomaly, utilizing angiocardiography to demonstrate absence of the right pulmonary artery, was reported a year earlier by Madoff et al. [*2*].

Recent reviews indicate that there are forty cases of absence of a main branch of the pulmonary artery recorded in the literature [*3,4*]. Of the total, about one-half (nineteen patients) had absence of the left pulmonary artery associated with tetralogy of Fallot; one patient reported by Nadas et al. [*5*] had absence of the right pulmonary artery and tetralogy of Fallot associated with dextrocardia and *situs inversus* (embryologically equivalent to absence of the left pulmonary artery with levocardia). McKim and Wigglesworth [*6*] reported six cases of absence of the left pulmonary artery and congenital heart disease; one case was associated with the Eisenmenger's complex (confirmed at necropsy). The remaining nineteen cases of isolated absence of a main branch of the pulmonary artery (most often the right) were in marked contrast because they were usually asymptomatic.

In the three cases reported herein isolated absence of the right pulmonary artery was associated with cystic bronchiectasis of the same lung in one patient. Lack of significant pulmonary fibrosis in the right lung in this case makes it unlikely that the absence of the right pulmonary artery was secondary to the bronchiectasis. In the second, an atrial septal defect was associated with absence of the left pulmonary artery. The last case, with absence of the right pulmonary artery, was associated with the Eisenmenger complex and a right pulmonary artery arising from the ascending aorta was noted.

ABSENCE OF RIGHT PULMONARY ARTERY WITH BRONCHIECTASIS RIGHT LUNG

CASE 1. A forty-one year old housewife (N. Y. H. #173743) was admitted on April 4, 1957, complaining of pain in the right chest. As a child the patient had frequent colds and upper respiratory infections; before the age of ten years she had had four bouts of pneumonia. Each winter she had had a severe cold with cough and expectoration lasting several weeks. Twenty years prior to admission many rales and rhonchi were heard over the right chest during a prenatal examination. On x-ray examination the heart and trachea were found to be in the right chest. In the ensuing twenty years she was seen many times in the clinic because of recurrent colds and cough; despite the abnormal physical signs at the right base, the x-ray of the chest had remained unchanged; the abnormal position of the heart was attributed to dextrocardia. A month before admission, she had an unusually severe cold with pain in the right posterior chest radiating around to the breast region, associated with slight cough and expectoration which for four days contained minute specks of blood. She denied dyspnea and weight loss.

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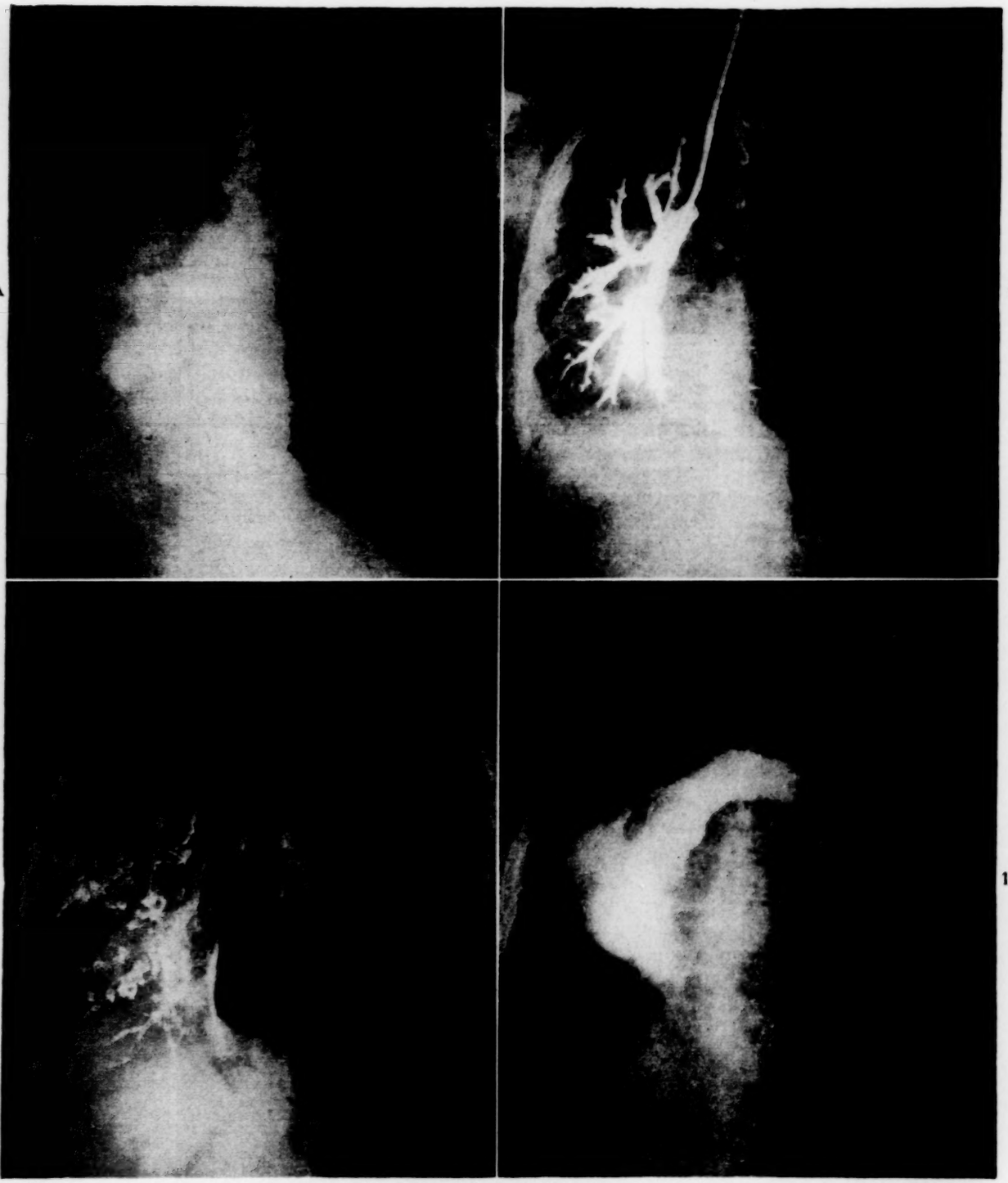


FIG. 1. CASE 1. A, frontal teleoroentgenogram shows deviation of the heart to the right. There is mediastinal herniation and the left lung is well aerated and vascular. The right lung is hypoplastic and contains a fine lacy pattern of blood vasculature at the periphery. B, frontal bronchogram shows the trachea deviated to the right; the right bronchial tree is distorted and uniformally dilated. C, bronchogram, at a later stage, reveals multiple and generalized cystic bronchiectasis of the right lung. D, angiocardiography shows absence of the right pulmonary artery. The heart is rotated in a mild (about 25 degree) left anterior oblique position.

On examination the patient was well developed and nourished and had a continuous non-productive cough. There was diminished expansion of the right chest. Dullness and many moist rales were heard posteriorly; the left lung was clear. The heart was percussed 1 cm. beyond the right sternal border. There were no murmurs; the blood pressure was 126/80 mm. Hg. Laboratory data showed: hemoglobin, 13.4 gm./100 cc.; hematocrit, 40 per cent and leukocyte count 12,300/cu. mm. Smears and sputum culture for acid fast bacillus were negative. The electrocardiogram was normal. Pulmonary function studies revealed a diminished vital capacity (70 per cent of normal); the maximum breathing capacity, however, was normal.

A roentgenogram of the chest (Fig. 1A) revealed the characteristic findings of absence of a right pulmonary artery. There was overdistension of the left hemithorax with deviation of the mediastinum, trachea and heart towards the right side. The volume of the right lung was decreased and the vasculature appeared meager. Bronchography (Fig. 1B and C) showed saccular bronchiectasis of the right lung with marked deformity of the right bronchial tree. Angiocardiography on April 9, 1957, established absence of the right pulmonary artery; the heart appeared somewhat dextrorotated and the left lung had overdistended pulmonary arterial vasculature. (Fig. 1D.) At the time of aortic filling there was filling of fine lacy vessels in the right lung characteristic of bronchial arteries. (Fig. 1E.)

The patient improved after bed rest and antibiotic therapy. Because of the small quantity of sputum and general good condition of the patient, further observation rather than pneumonectomy was advised.

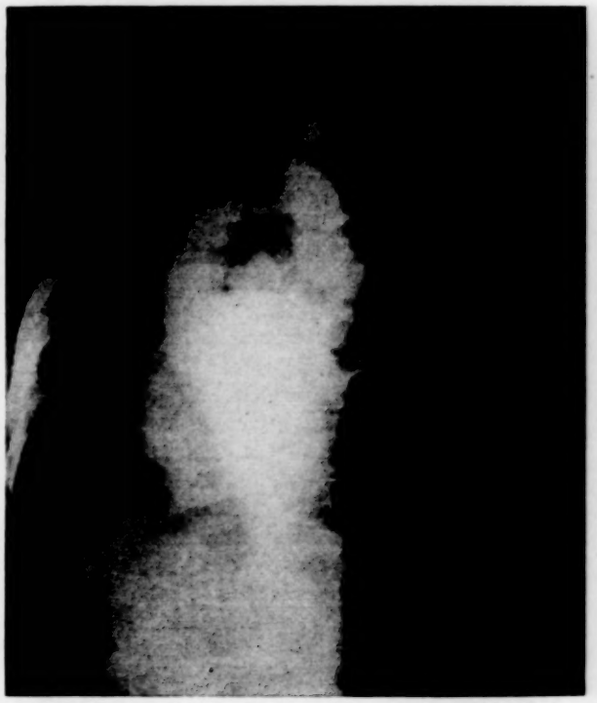


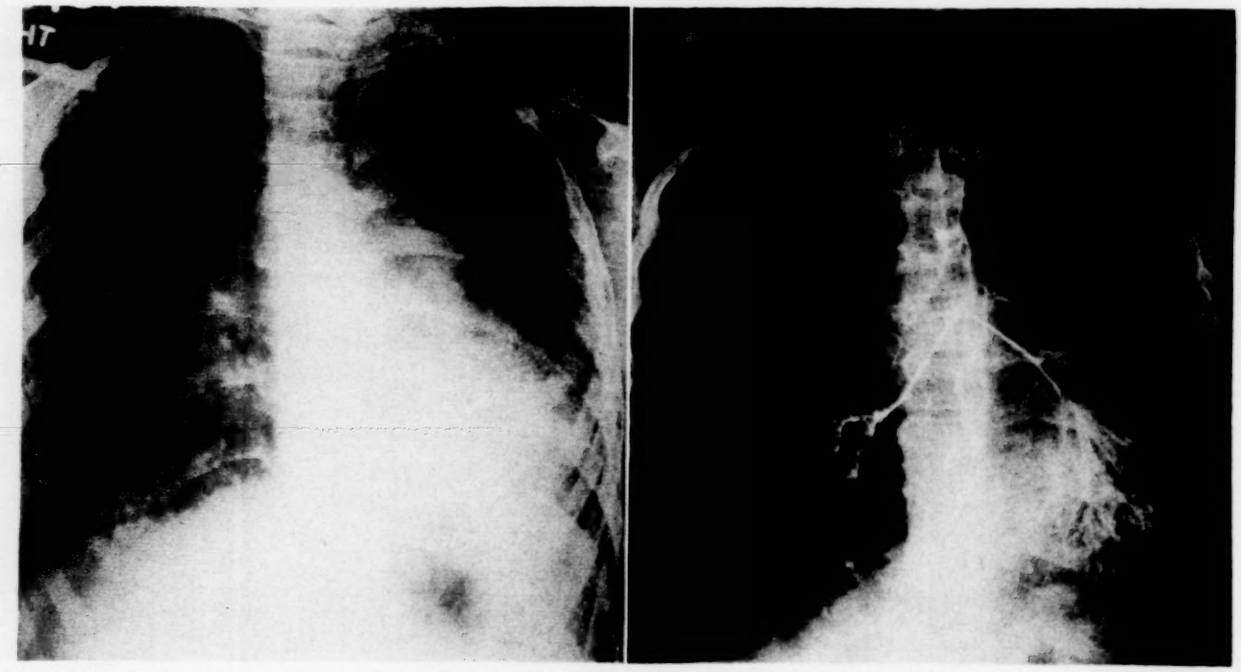
FIG. 1E. CASE I. When the structures in the left side of the heart are filled, there are pulmonary veins from the left lung; the right are absent. The aorta and brachiocephalic arteries are normal in position. Later studies revealed filling of the right pulmonary vasculature during the late stages of aortic opacification in a pattern characteristic of bronchial arterial circulation.

CONGENITAL ABSENCE OF THE LEFT PULMONARY ARTERY ASSOCIATED WITH ATRIAL SEPTAL DEFECT

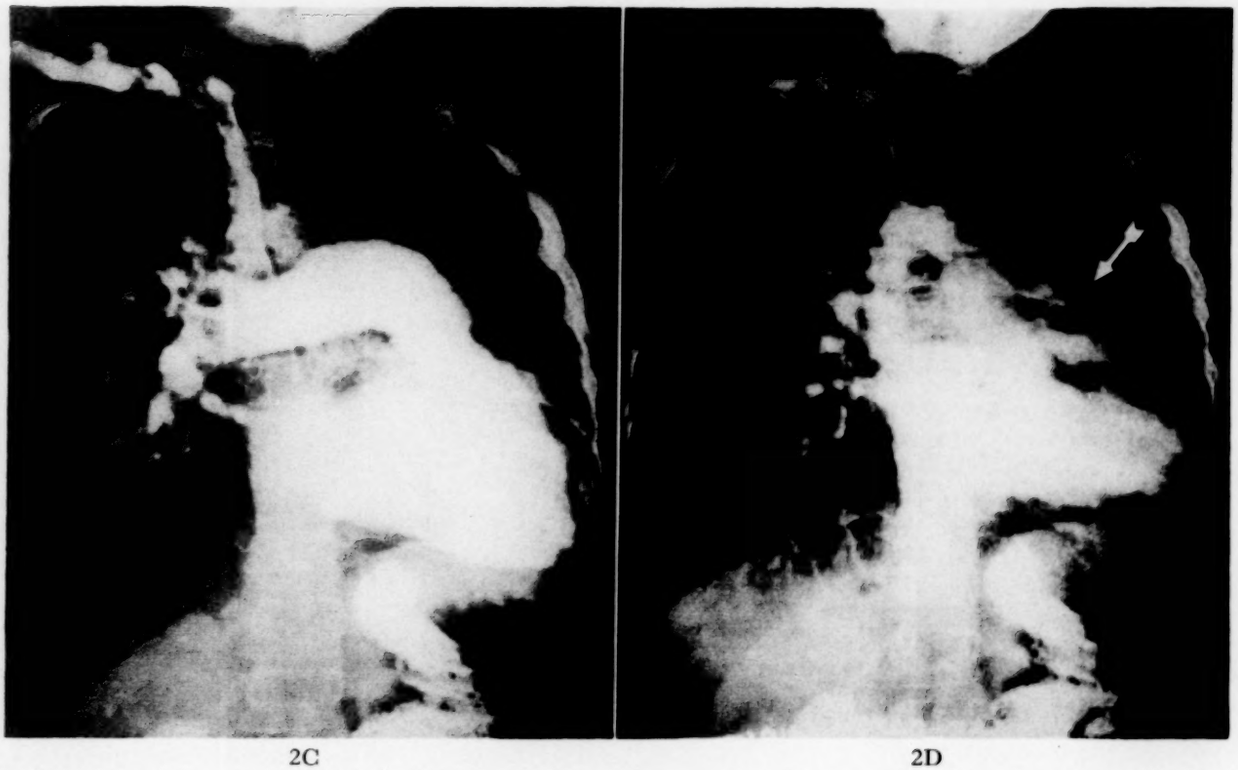
CASE II. A five and a half year old white girl (N. Y. H. #540101) was first seen in the clinic, June 6, 1949. She was born at full term; weight, 5 pounds, 13 ounces. Development was normal; at the age of five months bronchitis with fever and cough appeared; examination disclosed a heart murmur. From then on, until the age of three years, there were several attacks of acute upper respiratory infections associated with bouts of cyanosis. On examination a harsh systolic murmur, maximal at the base was heard. For the next six years, she visited the clinic periodically and remained well save for frequent upper respiratory infections. In January 1955, the patient had a persistent cough accompanied with mild cyanosis and temperature of 102°F. She was treated with broad spectrum antibiotics. When rales in the right lower lobe persisted, hospital admission was advised.

Physical examination on February 23, 1955, revealed a well developed and well nourished girl of eleven years, in no distress. There was a slight precordial bulge on the left. There was dullness and decreased breath sounds at the left base while at the right base there was dullness, increased tactile and vocal fremitus and bronchial breathing; there were no rales. The heart was enlarged, the position of maximal impulse was in the fourth interspace in the left mid-clavicular line. The apex impulse was strong and visible. No thrills were felt and the rhythm was normal sinus, rate 110. The second pulmonic sound was louder than the aortic. A harsh (grade 3) systolic murmur was heard all over the precordium, loudest in the third intercostal space to the left of the sternum. The blood pressure was 100/60 mm. Hg.

Laboratory data revealed: hemoglobin, 13.6 gm./100 cc.; erythrocytes, 5 m./cu. mm.; leukocyte count, 13,550/cu. mm. Electrocardiographic studies showed sinus tachycardia, a semivertical position of the heart and right ventricular hypertrophy with the right and left ventricles of approximately equal electrical importance. X-ray of the chest after subsidence of the fever (Fig. 2A) disclosed a plethoric right lung; the heart, trachea and mediastinum were rotated into the left hemithorax. Bronchography (Fig. 2B) revealed crowding of the left lower segmental bronchi. Angiocardiography (Fig. 2C to F) disclosed an absent left pulmonary artery. Right to left shunting of blood through a septal defect was also demonstrated. (Fig.

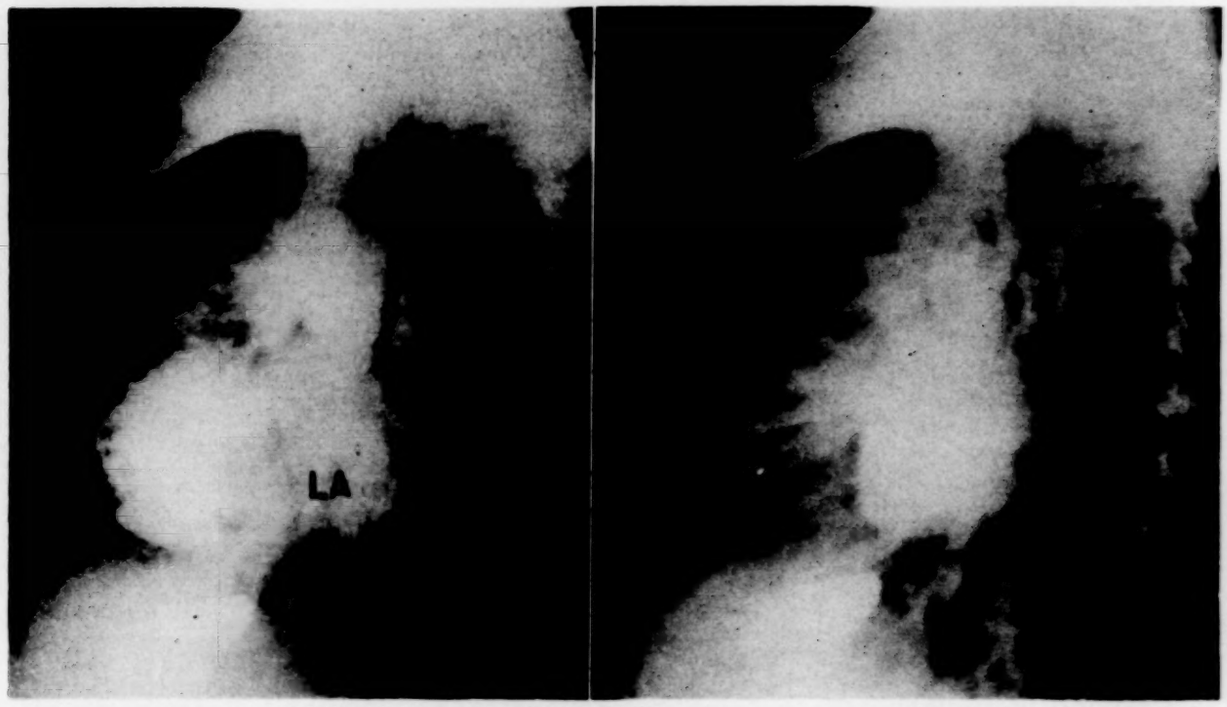


2A 2B



2C 2D

FIG. 2A to D. CASE II. A, frontal roentgenogram shows enlargement and rotation of heart and mediastinum into left hemithorax. There is plethora of the right pulmonary vasculature; the left lung pulmonary arterial circulation is absent. B, frontal bronchogram shows hypoplasia, crowding and distortion of the left lower bronchial tree. The right bronchus, although inadequately filled, is normal. C, frontal angiocardiogram reveals rotation of the heart in a right oblique fashion; there is absence of the left pulmonary artery. The right pulmonary artery and branches are enlarged. D, when the left side of the heart is opacified, the aorta is right-sided, the innominate artery is on the left and the common carotid and subclavian arteries are on the right side of the body. Persistence of right pulmonary arterial opacification (arrow) is due to a cardiac shunt.



2E 2F

FIG. 2E and F. Case II. E, left anterior oblique angiocardiogram demonstrates early (one and one-half seconds) opacification of the left atrium (LA) as well as of the structures in the right side of the heart, pulmonary artery and right branch; evidence of the right-left shunt. F, left anterior oblique angiocardiogram shows opacification of the left atrium, ventricle and aorta. Note persistent opacification of the right pulmonary artery indicating a left-right shunt.

2E.) At the time of aortic filling reopacification of the right cardiac chambers and pulmonary artery was seen, indicating left-to-right blood flow (Fig. 2D and F); there was also a fine lacy network of blood vessels in the left lung having the appearance of bronchial arterial vessels. After treatment with antibiotics the patient recovered and was asymptomatic. When last seen in the clinic on March 1, 1957, she had gained weight and remained free of respiratory infections but complained of fatigue in the afternoon.

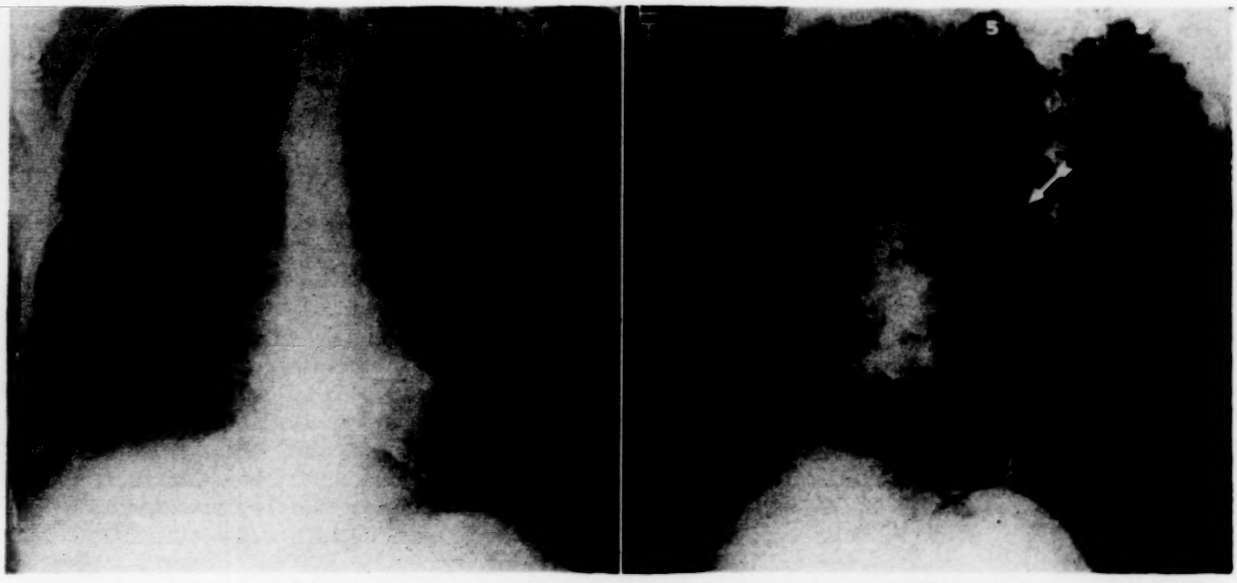
CONGENITAL ABSENCE OF THE RIGHT PULMONARY ARTERY ASSOCIATED WITH THE EISENMENGER COMPLEX AND RIGHT PULMONARY ARTERY FROM THE ASCENDING AORTA

CASE III. A twelve year old schoolgirl was admitted to St. Francis Hospital and Sanatorium on May 17, 1956, because of dyspnea, cyanosis and easy fatigability on exertion. At the age of five and a half months, a cardiac murmur was heard. She had had three attacks of pneumonia and was dyspneic and mildly cyanotic on exertion. On examination the patient was well developed and nourished. The lungs were clear. A systolic (grade 2) murmur was heard in the fourth left interspace and there was mild clubbing and cyanosis of the fingers. The blood pressure was 92/70 mm. Hg. Laboratory data revealed: hemoglobin, 14.0 gm./100 cc.; erythrocytes, 4.6 m./cu. mm. and leukocytes 7,900/cu. mm. The electrocardiogram showed normal sinus rhythm with sinus arrythmia, right ventricular hypertrophy, right axis deviation and vertical heart position. X-rays of the chest (Fig. 3A to C) showed moderate enlargement of the heart; the vasculature of the right lung was markedly decreased. The left anterior oblique film showed undue prominence of the left pulmonary artery. (Fig. 3B.) Cardiac catheterization revealed markedly increased right ventricular (60/3) and pulmonary artery (60/22 mm. Hg) pressures with increased right heart oxygen saturations due to a high ventricular septal defect. The pulmonary vascular resistance of the left lung was moderately increased.

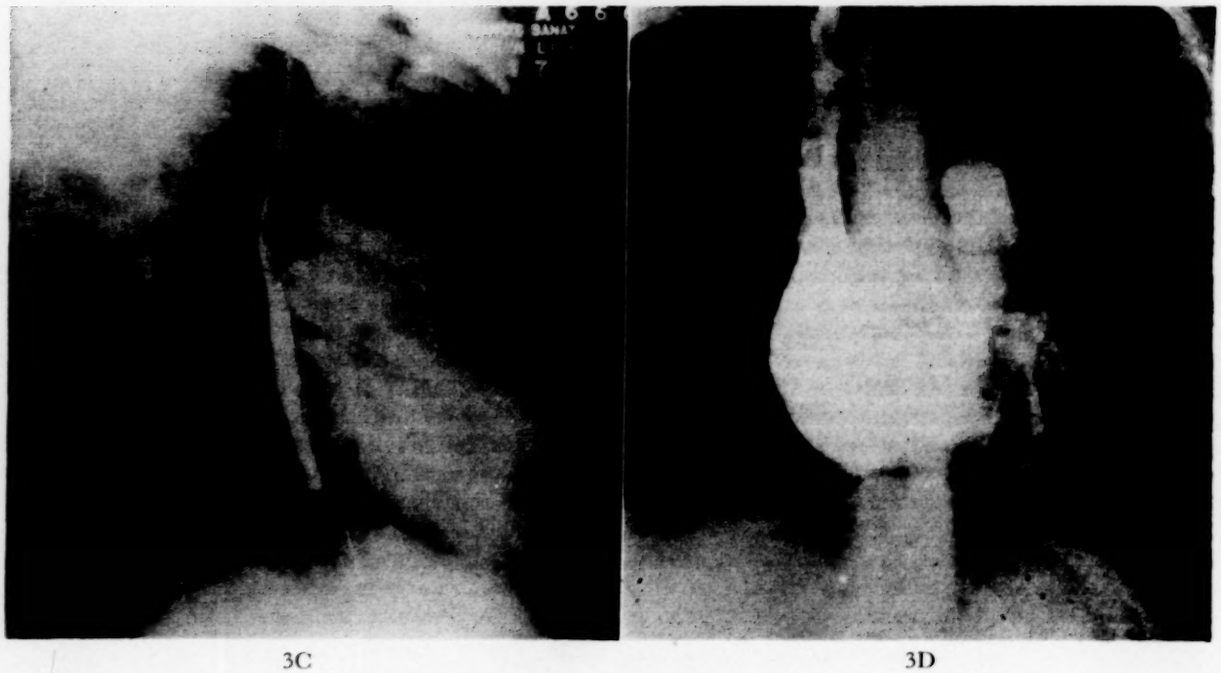
Angiocardiography (Fig. 3D and E) disclosed an absent right pulmonary artery. Aortic filling was seen early suggesting an over-riding aorta but maximum opacification of the aorta occurred at the time of left heart filling. Pulmonary veins from the left lung draining into the left atrium were clearly recognized and there was also reopacification of the pulmonary artery and left branch at the time of left heart filling. (Fig. 3E.) A small vessel giving off ascending and descending branches was seen arising from the mid-ascending aorta and supplied the right lung, constituting a pulmonary arterial circulation from the ascending aorta. (Fig. 3E.)

COMMENTS

There appear to be three main types of congenital absence of a main branch of the pul-



3A 3B



3C 3D

FIG. 3A to D. CASE III. A, frontal roentgenogram shows heart enlargement. Left pulmonary artery and tree appear normal. The right pulmonary vasculature is diminished. The right lung is slightly smaller than the left; the heart and mediastinal structures are in the mid-line. B, left anterior oblique position shows a left pulmonary artery (arrow); the right cannot be seen. C, the right anterior oblique esophagram suggests absence of the right pulmonary artery. D, frontal angiocardiogram showing absence of a right pulmonary artery.

monary artery. The first, consists of an isolated absence of a pulmonary artery (most often the right) unassociated with a cardiac septal defect, often accompanied with a high incidence of aortic anomalies. Rarely, as in Case I, may bronchiectasis of the lung with the absent pulmonary artery occur. In the third group, cardiac septal defects, most often a high ventricular septal defect and infundibular stenosis (tetralogy of Fallot) associated with absence of the left pulmonary artery occur. Absence of the left pulmonary artery associated with the Eisenmenger complex has also been described in another case [*6*]. In Case III, however, the

Eisenmenger's complex was associated with absence of the right pulmonary artery. In Case II, an atrial septal defect was associated with absence of the right pulmonary artery.

Although many attempts have been made to explain the absence of a main branch pulmonary artery on embryologic grounds [*2,4,6*]; none are entirely satisfactory. There is general agreement that the pulmonary artery arises from the sixth aortic arch, at the same time, or soon after the development of the fourth arch which eventually becomes the aortic arch of the adult. The right and left pulmonary arteries do not develop symmetrically but seem to have separate embryologic origins [*4,7*]. Absence of the left pulmonary artery is commonly associated with the tetralogy of Fallot [*4–6*]. A right-sided aortic arch, present in 20 per cent of patients with tetralogy of Fallot, is found in 60 per cent of the cases with associated absence of the left pulmonary artery [*4*]. In the cases of isolated absence of a pulmonary artery without a cardiac shunt, the right pulmonary artery is usually absent. Associated lesions, when present, consist of aortic anomalies: a right-sided aorta; patent ductus arteriosus; or coarctation of the aorta [*4*].

Bronchiectasis in the lung with absence of a pulmonary artery (Case I) is rare. In 1946, a thirty-one year old man with extensive cystic bronchiectasis of the left lung was studied [*8*]; the absent left pulmonary artery was thought to be secondary to the pulmonary disease. Its resemblance to Case I, although the left lung was bronchiectatic, suggests that it may be an example of congenital absence of the left pulmonary artery complicated by bronchiectasis. Another woman, fifty-three years of age, with absence of the left pulmonary artery and bronchiectasis of the left lung was studied recently at Kings County Hospital, Brooklyn [*9*]. Whether or not the bronchiectasis in these cases is of congenital origin is unknown; it appears more likely that they are acquired.

Usually, the systemic blood supply of the lung with the absent pulmonary artery is derived from bronchial arteries. This can be suspected by the fine lacy pattern of the pulmonary vasculature [*10*] and may be verified by their opacification soon after aortic filling. In Case III, a pulmonary artery was demonstrated arising from the ascending aorta. (Fig. 3E.) A similar case associated with absence of the right pulmonary artery complicated by a patent ductus arteriosus but with an intact ventricular septum was recently described; an attempt to connect the systemic right pulmonary artery to the main stem pulmonary artery via a plastic graft after ligation of the ductus was not successful [*11*].

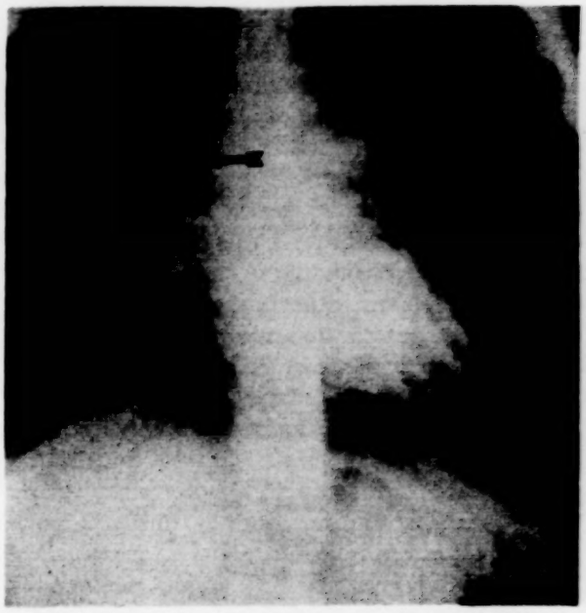


FIG. 3E. Case III. E, angiocardiogram at six seconds shows left atrial, left ventricular and aortic opacification. Left pulmonary artery, branches and pulmonary veins are contrasted. Note upper and lower systemic pulmonary arteries arising from ascending aorta (arrow).

Characteristic physical and roentgen findings are present when there is absence of a main branch pulmonary artery unassociated with bronchiectasis or intracardiac septal defects. There is a lag in expansion and diminution of the breath sounds on inspiration in the lung with the absent pulmonary artery. Roentgenography reveals one of the lungs to be small and containing fine vascular markings usually at the periphery of the lung. The opposite lung is overdistended, well vascularized and displaces the heart, trachea and mediastinal structure producing mediastinal herniation. Both lungs are clear, well aerated and free of disease. Bronchography reveals that the bronchus of the lung with the absent pulmonary artery is patent and normal in appearance [*3,12*]. Angiocardiography is diagnostic and demonstrates absence of the pulmonary artery in the hypoplastic lung. Angiocardiographic study of the pulmonary vasculature of the lung with the absent pulmonary artery also demonstrates that opacification follows aortic filling, the pattern conforming to the bronchial arterial circulation.

Patients with the isolated and uncomplicated type of congenital absence of a pulmonary artery are usually asymptomatic [*1–3,13,14*]. One patient, however, complained of hemoptysis which was attributed to the hyperemia produced by the bronchial arterial circulation [*15*]. With bronchiectasis, although mediastinal shift is quite evident, there may be further alteration of the pulmonary parenchyma due to fibrosis and cystic changes. A history of recurrent infections, cough and expectoration and the finding of rales are in marked contrast to the asymptomatic group with isolated absence of a pulmonary artery branch. Bronchography establishes the diagnosis of bronchiectasis. (Fig. 1B and C.)

Diagnosis of absence of a pulmonary artery when there are cardiac septal shunts is more subtle. Recurrent pulmonary infections, bouts of cyanosis and dyspnea are manifestations of abnormal shunting of the circulation, which is readily recognized by cardiac murmurs. Conventional roentgenography in this group, in contrast to the isolated type with absence of a pulmonary artery, provides scant clues for diagnosis. In the patient with the atrial septal defect (Case II) rotation of the heart into the left hemithorax obscured the absent pulmonary arterial circulation of the left lung (Fig. 2A); angiocardiography was essential for diagnosis of absence of the left pulmonary artery. (Fig. 2C and E.) Bronchography established the presence of an intact, although crowded left bronchial tree (Fig. 2B) and excluded agenesis of the left lung [*16*]. Diagnosis of absence of the right pulmonary artery in Case III was even more difficult; the heart appearing in the midline and mediastinal herniation being absent. Furthermore, only slight differences in the vasculature of the two lungs were present. (Fig. 3A.) Angiocardiography was essential because the systemic pulmonary artery from the ascending aorta obscured the absence of the right pulmonary artery. The systemic pulmonary artery apparently also prevented hypoplasia of the right lung, thereby, making unavailable an important clue for diagnosis of absence of a pulmonary artery branch by conventional roentgenography.

Bronchospirometric [*2,3,15*], pulmonary function [*1,2,15*], and cardiac catheterization [*1,15*] studies have been performed in patients with the isolated types of absence of a pulmonary artery. They confirm that such patients have only mild functional disability. Lung volumes, maximum breathing capacity, ventilation and blood gas determinations were normal [*1,2,15*]. One patient had a moderately reduced vital capacity [*15*]. Bronchospirometry studies, as expected, showed a marked difference in oxygen uptake, being practically negligible on the side with the absent pulmonary artery when pure O_2 was substituted for air [*3*]. Another group of investigators reported 7 per cent O_2 uptake when pure O_2 was substituted for air [*2*], presumably due to an increased uptake from the bronchial arterial system. Cardiac catheterization showed normal to mild elevations of the pulmonary artery pressures [*1*]. This is entirely in keeping with the findings reported after pneumonectomy and ligation of a pulmonary artery [*1*].

Patients with isolated absence of a pulmonary artery should have prompt treatment of respiratory infection. Case II, with an atrial septal defect, illustrates the severe disability that results when infection occurred in the "good lung." Whether or not pneumonectomy should be performed for bronchiectasis involving the lung with absence of pulmonary artery will depend upon the general condition of the patient and the severity of the symptoms. Overdistention of the vascular lung occurs regularly with absence of a pulmonary artery. Whether or not such a patient will be able to adjust to the altered thoracic dynamics after pneumonectomy is unknown. In the absence of serious disability caused by bronchiectasis, a conservative rather than an aggressive surgical approach seems warranted. Other authors have commented on the dangers of doing a Blalock or Potts operation for relief of the anoxia in Fallots' tetralogy when absence of the left pulmonary artery is unrecognized [*4–6*]. However, when the mortality of open heart surgical correction of ventricular septal defects and infundibular stenosis (Fallots' tetralogy) becomes lower, it is reasonable to expect that such patients may be benefited by corrective surgery. Study of patients with the isolated types of absence of a pulmonary artery has indicated that good health may be anticipated when there is congenital absence of a pulmonary artery. Present day surgical technics are available for the correction of atrial septal defects. However, surgery should be preceded by cardiac catheterization in order to establish the degree of pulmonary arterial shunting and pressure and to exclude the septum primum type of atrial defect. The patient with the Eisenmenger's complex (Case III) is probably inoperative because of the increased pulmonary resistance in the left lung.

SUMMARY AND CONCLUSIONS

Congenital absence of a main branch of the pulmonary artery may be classified in two categories; isolated, and associated with septal defects. Unless there is bronchiectasis in the lung with the absent pulmonary artery, patients in the first group are usually asymptomatic. Pulmonary function and hemodynamic studies in the isolated types are usually within the normal range, even though oxygen uptake on the side with the absent pulmonary artery is negligible. Diagnosis of congenital absence of a pulmonary artery may often be made by inspection of the conventional roentgenogram which shows a hypoplastic poorly vascularized lung on one side with a fully vascularized distended opposite lung that causes mediastinal herniation. In contrast, patients with cardiac septal defects and absence of a pulmonary artery are often ill with recurrent infections, dyspnea and cyanosis. The left pulmonary artery is usually absent and diagnosis from study of the conventional roentgenogram is difficult.

Angiocardiography provides the definitive diagnosis of absence of a pulmonary artery main branch. Cardiac catheterization may be needed to establish the level of a cardiac shunt, whether atrial or ventricular; in the latter, it is also necessary to differentiate between the tetralogy of Fallot and the Eisenmenger's complex. Angiocardiography also provides information regarding the systemic circulation in the lung with the absent pulmonary artery. Indirect evidence of bronchial arterial circulation can be demonstrated by the circulatory pattern following aortic opacification; ascending aortic pulmonary arterial branches, if present, can readily be recognized. Differentiation of the isolated from the complicated types of absence of a pulmonary artery is significant because of therapeutic and prognostic implications.

ADDENDUM

Recently, Alley and associates [*17*] have reported absence of filling of the left pulmonary artery in the angiocardiogram of a patient with long-standing suppurative disease of the left lung. Retrograde filling of the left pulmonary artery via huge bronchial arterials were demonstrated by aortography. Hemodynamic and electrocardiographic data also supported the presence of reversal of blood flow from the systemic (bronchial) to the lesser (pulmonary) circulations. Their findings, especially the demonstration of huge bronchial arterial collaterals, are evidence of functional rather than congenital absence of a main branch of a pulmonary artery such as is described in Case 1.

REFERENCES

1. Steinberg, I., Dotter, C. T. and Lukas, D. S. Congenital absence of a main branch of the pulmonary artery. *J. A. M. A.*, 152: 1216, 1953.
2. Madoff, I. M., Gaensler, E. A. and Streider, J. W. Congenital absence of the right pulmonary artery: diagnosis by angiocardiography with cardiorespiratory studies. *New England J. Med.*, 247: 149, 1952.
3. Smart, J. and Pattinson, J. N. Congenital absence of left pulmonary artery. *Brit. M. J.*, 1: 491, 1956.
4. Emanuel, R. W. and Pattinson, J. N. Absence of the left pulmonary artery in Fallot's tetralogy. *Brit. Heart J.*, 18: 289, 1956.
5. Nadas, A. S., Rosenbaum, H. D., Wittenborg, M. H. and Rudolph, A. M. Tetralogy of Fallot with unilateral pulmonary atresia; a clinically diagnosable and surgically significant variant. *Circulation*, 8: 328, 1953.
6. McKim, J. S. and Wigglesworth, F. W. Absence of the left pulmonary artery: a report of six cases with autopsy findings in three. *Am. Heart J.*, 47: 845, 1954.
7. Bremer, J. L. On the origin of the pulmonary arteries in mammals. *Anat. Rec.*, 3: 334, 1909.
8. Steinberg, I., Dotter, C. T. and Andrus, W. D. Angiocardiography in thoracic surgery. *Surg., Gynec. & Obst.*, 90: 45, 1950.
9. Lyons, H. A. Personal communication.
10. Campbell, M. and Gardner, F. Radiological features of enlarged bronchial arteries. *Brit. Heart J.*, 12: 183, 1950.
11. Caro, C. G., Lermando, V. C. and Lyons, H. A. Aortic origin of the right pulmonary artery. *Brit. Heart J.*, 19: 345, 1957.
12. Steinberg, I. and Finby, N. Clinical and angiocardiographic features of congenital anomalies of the pulmonary circulation: a classification and review. *Angiology*, 7: 378, 1956.
13. Wyman, S. W. Congenital absence of a pulmonary artery: its demonstration by roentgenography. *Radiology*, 62: 321, 1954.
14. Maier, H. C. Absence or hypoplasia of a pulmonary artery with anomalous systemic arteries to the lung. *J. Thoracic Surg.*, 28: 145, 1954.
15. Flynn, J. E., Siebens, A. A. and Williams, S. F. Congenital absence of a main branch of the pulmonary artery. *Am. J. M. Sc.*, 228: 673, 1954.
16. Lukas, D. S., Dotter, C. T. and Steinberg, I. Agenesis of the lung and patent ductus arteriosus with reversal of flow. *New England J. Med.*, 152: 1216, 1953.
17. Alley, R. D., Stranahan, A., Kausel, H., Formel, P. and Van Mierop, L. H. S. Demonstration of bronchial-pulmonary artery reverse flow in suppurative pulmonary disease. *Clin. Research*, 6: 41, 1958.

Review

Coccidioidomycosis and Its Treatment with Amphotericin B*

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Recognition of systemic mycotic disease in man has been simplified considerably during the past decade by many improvements in diagnostic methods. The true incidence of the systemic mycoses, however, is not accurately reflected in the total number of cases reported in the world medical literature. Attention has been drawn to the fact that, while a total of only 300 cases of cryptococcosis have been reported in the world literature to date, 151 fatal cases were recorded in the United States alone in the four-year period following 1949 [*1*]. Despite improvements in diagnostic procedures, recognition of systemic mycotic disease still is governed more by the availability of laboratory personnel experienced in the required technical procedures than by any other factor.

Systemic mycotic infections discovered in hospital centers in the United States fall into one of the following categories: (1) Primary infections, such as coccidioidomycosis, histoplasmosis, North American blastomycosis, cryptococcosis, moniliasis, sporotrichosis, actinomycosis and nocardiosis. The incidence of the first three infections varies with the geographical regions of the country. (2) Secondary infections, usually clinical variations of moniliasis. These emerge as complications during the treatment of bacterial or viral infections with broad spectrum antibiotics. Cases of this type are now fairly numerous and have a uniform geographic distribution. (3) Secondary infections, such as cryptococcosis, histoplasmosis, mucormycosis, aspergillosis and moniliasis. These appear as complications in persons suffering from primary debilitating diseases such as Hodgkin's disease, lymphosarcoma, leukemia, sarcoidosis, tuberculosis or other primary mycoses, and emerge especially among those who may have received treatment with corticosteroids, nitrogen mustard or antibiotics. In many instances fatalities ensue from invasion of the tissues by the pathogenic fungi rather than as a result of the primary illness.

One of the most important of the primary systemic mycoses, particularly to residents and visitors of the southwestern and western regions of the United States, is coccidioidomycosis. The etiologic agent of this disease is Coccidioides immitis (Figs. 1 to 4), a hardy filamentous mold which thrives in the soil of relatively arid regions in California, New Mexico, Arizona, Texas, Nevada and Utah, and in the northern states of Mexico. Infection is usually acquired by inhalation of tiny arthrospores of the fungus. In endemic areas these are believed to be dispersed into the air by winds, storms and other disturbances of infected soil and vegetation.

The highly infectious nature of coccidioidomycosis is illustrated by the fact that even limited exposure to infected dust during brief travel through an endemic area may be sufficient to cause infection. There is also a high incidence of laboratory-acquired infections, as is well known. On the basis of skin testing with coccidioidin, it is estimated that in endemic areas 10 to 15 per cent of newly arrived residents contract the infection within the first year and approximately 90 per cent within ten years [*2*]. This may occur with or without obvious clinical symptoms. It is estimated that millions of persons residing in and visiting the southwestern states have become infected with this fungus [*3*]. During World War II, coccidioidomycosis assumed considerable

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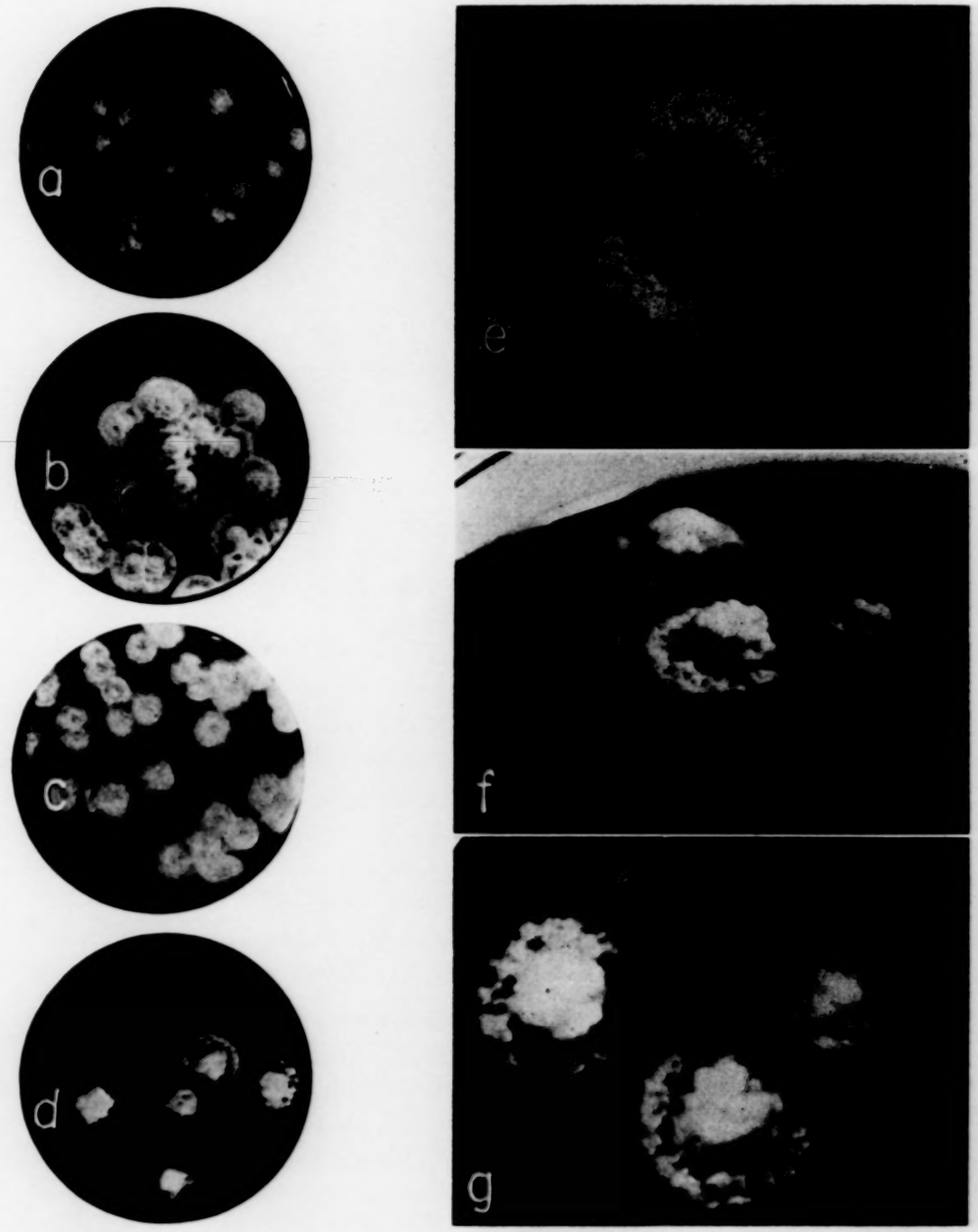

FIG. 1. Colonies of C. immitis on agar culture media. At first the colony is membranous and adherent, then it rapidly becomes covered with an abundant cottony white aerial mycelium which may become tan with age. (*a*) Brain-heart infusion blood agar, three days at 37°C., × ½. (*b*) Francis' cystine glucose blood agar, three days at 37°C., × ½. (*c*) Littman's liver–spleen glucose blood agar, three days at 37°C., × ½. (*d, f, g*) Littman oxgall agar, eight days at 20°C. Colonies show moist, membranous peripheries and cottony centers (*d*, × ½) (*f, g*, × 2). (*e*) Sabouraud dextrose agar, original size of giant colony, eight days at 20°C. Note central tuft of hyphae surrounded by radially furrowed sparse growth which is bordered by wide zone of dense white mycelium. AFIP Acc. No. 218120-C159.

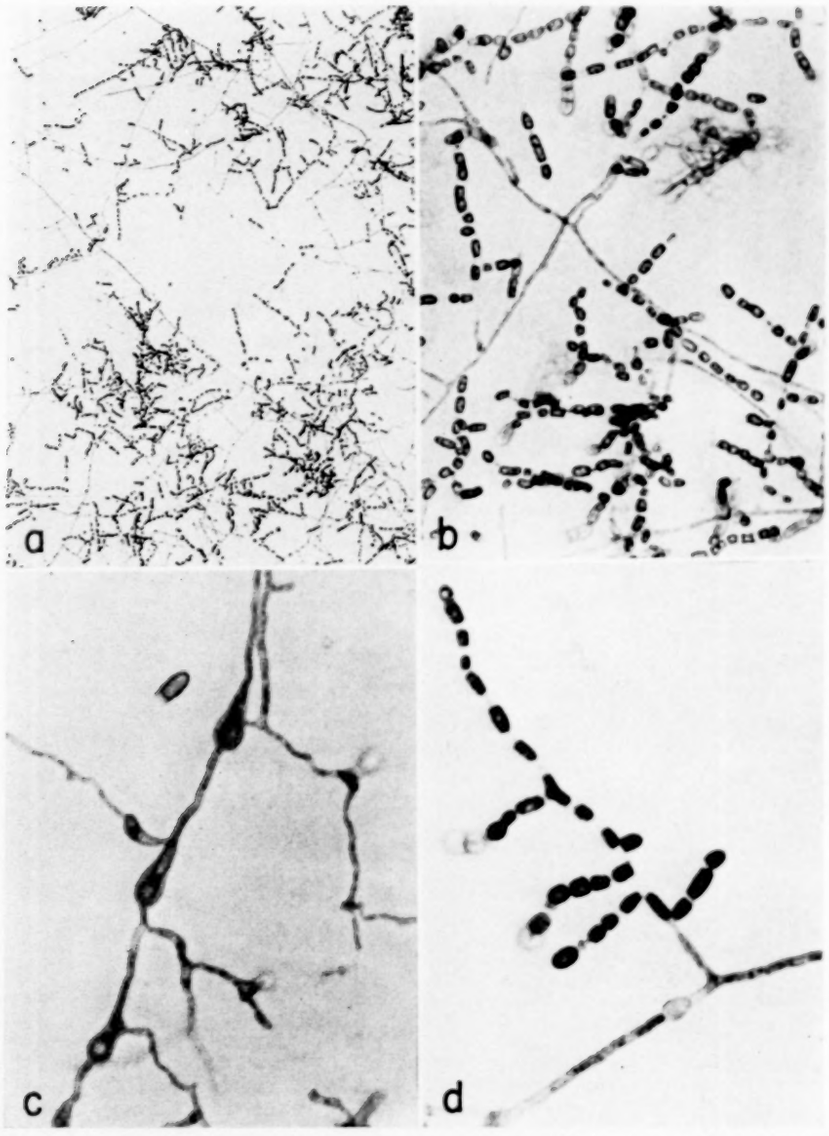

FIG. 2. Microscopic appearance of C. immitis in a Littman microculture (Fisher Scientific Co.) potato-dextrose agar at 30°C., lactophenol cotton-blue mount. (*a*) Abundant arthrospore formation, × 100. (*b*) Chains of characteristic rectangular arthrospores, some of which may be ellipsoidal or oval, separated by clear spaces, × 435. (*c*) Characteristic branching, septate mycelium, bearing racquet-shaped cells, × 805. (*d*) Specialized, thickened branches of aerial mycelium develop into chains of rectangular arthrospores which remain connected by hyphal remnants. These fragile connections are very easily parted and the arthrospores readily dispersed into the air, × 805. Inhaled infective arthrospores are promptly phagocytized and enter into a parasitic life cycle in man, see Figures 3 and 4. AFIP Acc. Nos. 218120-15, 243, 246 and 245.

epidemiological importance because of the large number of clinical infections (6,000) incurred by troops stationed in the southwestern region [*4*]. Despite this high morbidity, the mortality rate for this country is low, i.e., in 1955 only sixty-two deaths due to coccidioidomycosis were recorded by the National Office of Vital Statistics [*5*]. The same region of the United States is also enzootic for animals. Maddy [*6*] reported 3,173 cases of coccidioidomycosis in cattle which had acquired their infections in various parts of California, Arizona, New Mexico, Colorado and Texas. Maddy also described infection in sheep from California [*7*].

For all practical purposes the diagnosis of coccidioidomycosis may be excluded in persons who have neither visited nor resided in the endemic states listed. In rare cases, however, spread to non-endemic areas by fomites has been recorded. The movement of people be-

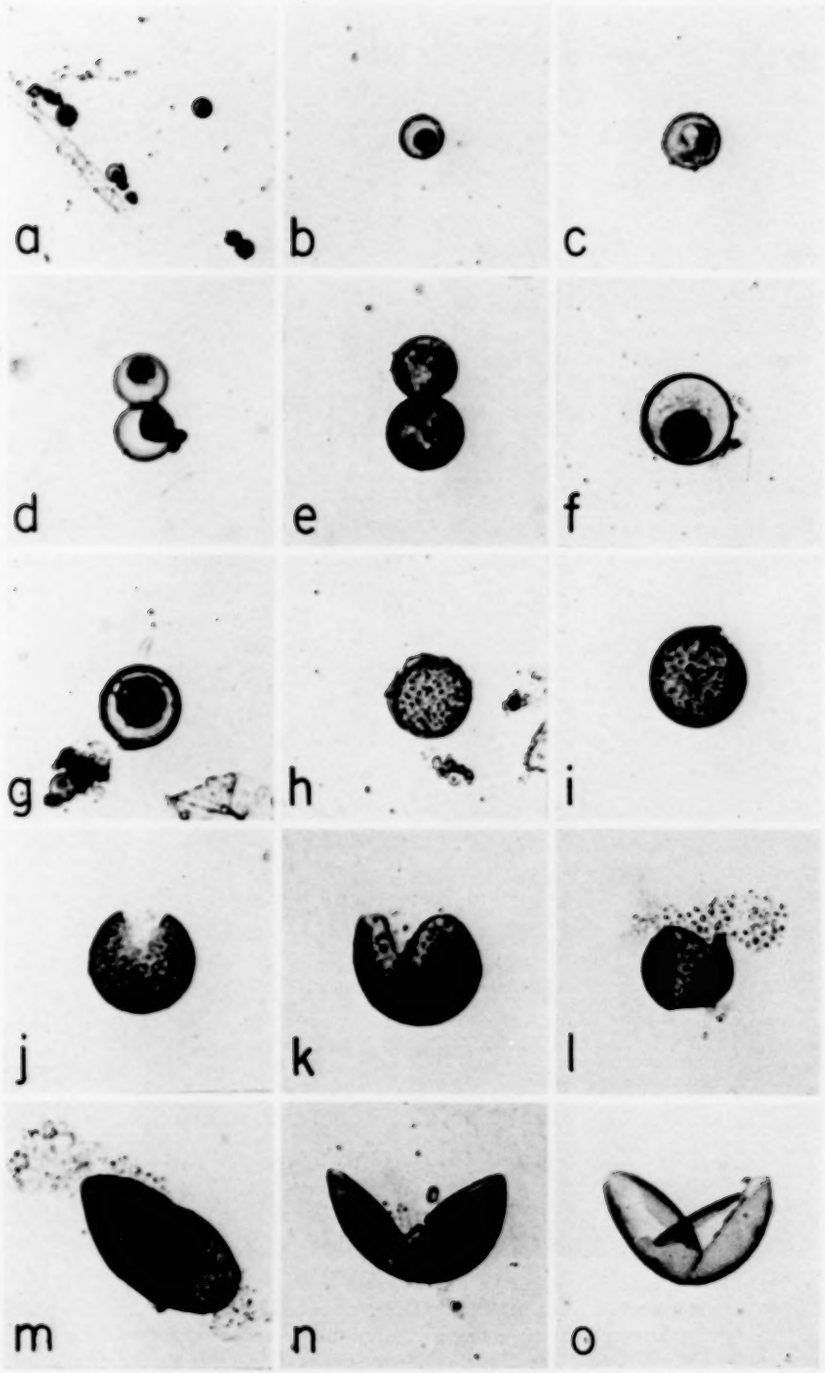

FIG. 3. Parasitic life cycle of C. immitis in man, from purulent abscess material, lactophenol cotton-blue mount. (*a*) Young developing spherules containing single nuclei, × 100. (*b–f*) Spherules increase in size with growth, some remain attached, × 435. (*g*) Beginning of endosporulation, × 435. (*h, i*) Fully matured spherule crowded with endospores, × 435. (*j–m*) Rupture of the spherule and liberation of endospores each of which becomes a new developing spherule, × 435. (*n, o*) Emptying of spherule, leaving sac, × 435. A spherule may vary in diameter from 10 to 80 microns and an endospore from 2.5 to 5 microns. AFIP Acc. No. 218120 series.

tween endemic and non-endemic areas in the United States removes coccidioidomycosis from the category of regional diseases and greatly widens its distribution. The disease may be overlooked in non-endemic regions because of failure to elicit a history of travel and residence in the highly endemic areas, or failure to perform mycological and serological studies or tests for

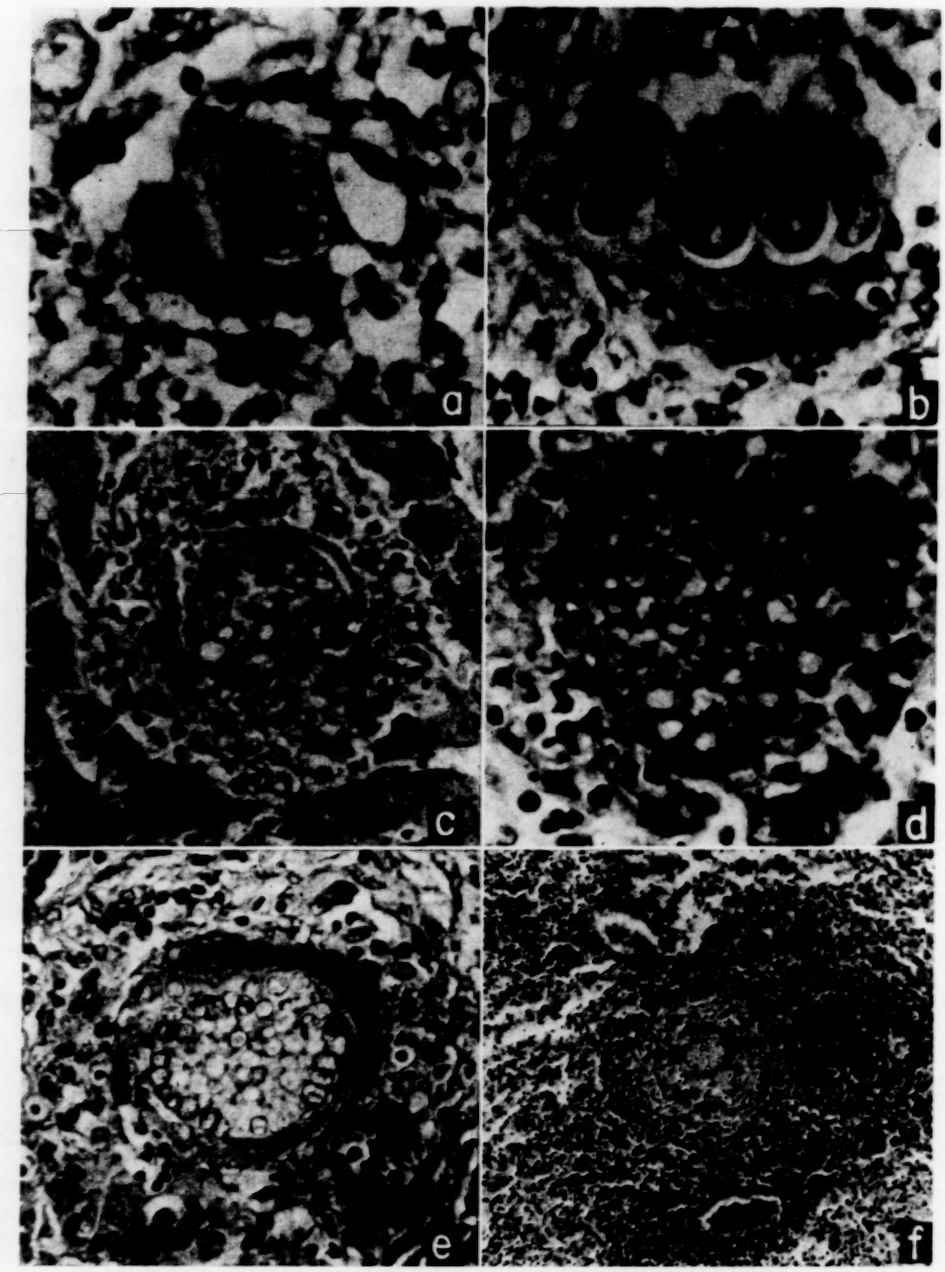

FIG. 4. Parasitic life cycle of C. immitis in tissues of man, hematoxylin and eosin stain. (*a*) Liberation of endospores within a giant cell, by a matured spherule, lung, × 805. (*b*) Released endospores, now young spherules, enlarging within a giant cell, lung, × 805. (*c*) Enlarging spherules, free in tissues, incite an epithelioid cell reaction and cause infiltration of polymorphonuclear leukocytes, liver, × 435. (*d*) Enlarging spherules grow away from their original entrapment, liver, × 805. (*e*) A fully matured spherule, with endospores, ready to rupture, has completely replaced the cytoplasm of a giant cell. Each endospore, upon release as a young spherule, begins a new cycle, lung, × 805. (*f*) Epithelioid cell reaction, which is attributed to phospholipid-rich spherule wall [*67*], is surrounded by heavy infiltration of polymorphonuclear leukocytes, same as (*e*), lung, × 100. AFIP Acc. Nos. 218120-379, 381, 374, 378, 386 and 385.

skin hypersensitivity. This is illustrated in the case histories that follow.

Coccidioidomycosis varies from an asymptomatic or mild influenza-like pulmonary infection (Valley Fever) to a fatal, progressive and disseminated granulomatous disease (coccidioidal granuloma). In a study of 1,351 primary coccidioidal infections of military personnel, Smith and co-workers [*8*] observed that in approximately 40 per cent symptoms of the dis-

ease developed, while 60 per cent were symptom-free. The latter group was detected by the emergence of skin hypersensitivity to coccidioidin and by the appearance of a non-specific type of pulmonary infiltration in roentgenograms of the chest. Generalized dissemination of the disease occurs in one of 400 white infected males (0.25 per cent), and in one of 2,000 infected females. The rate of dissemination in Negro males is approximately ten times higher than in white males. Coccidioidal infection occurs four times more frequently in men than in women [*9*], while erythema nodosum and erythema multiforme, the skin manifestations of the disease, occur twice as frequently in women as in men, and rarely in the Negro. The disease is more severe in Negroes, who usually require considerably longer hospitalization [*10*]. Man-to-man transmission has not been recorded.

The experience of Smith et al. [*11*] with 39,500 serological tests for coccidioidomycosis indicates that simultaneous precipitin and complement fixation tests of a patient's serum can provide a specific diagnosis of the disease, although occasional inconsistent cross-reactions with other mycoses do occur. In primary coccidioidomycosis, precipitins, which appear early in the blood serum, begin to decrease by the third week after infection and are no longer of diagnostic value by the fifth month after infection. The complement fixation titer of the serum, on the other hand, rises with the severity and duration of the infection. Values of 1:16 and 1:32 are indicative of dissemination, whereas subsiding titers indicate a favorable prognosis.

Skin hypersensitivity in coccidioidomycosis is ascertained by the intracutaneous injection of 0.1 ml. of 1:100 dilution of coccidioidin (Cutter Laboratories), usually made in parallel with other fungal antigens, such as blastomycin 1:1000 and histoplasmin 1:1000. A positive skin test indicates previous infection with C. immitis and is subject to the same limitations in interpretation as the tuberculin skin test. Patients with disseminated coccidioidomycosis usually show slight or negative reactions to 1:100 and 1:10 dilutions of coccidioidin.

Judging from the reviews of Kritzer, Biddle and Kessel [*12*], Schwarz and Muth [*13*] and Baum and Schwarz [*14*], an extensive literature on coccidioidomycosis has been accumulated. For more detailed discussions of this disease the reader is referred to the studies on pathologic aspects by Forbus and Bestebreurtje [*4*], the clinical and epidemiological reports of Smith and co-workers [*2,8,15–25*] and others [*10,11,26,27*], and to papers on surgical management [*28–32*], orthopedic problems [*33–37*], roentgen diagnosis [*33,38,39*], pediatric aspects [*36,40–42*] and microbiological studies [*43–47*].

Treatment of Coccidioidomycosis. Until the present time, the primary treatment of patients with coccidioidomycosis consisted of prolonged bedrest, the same method of treatment employed in the pre-antibiotic era for patients with pulmonary tuberculosis. The non-disseminated form of coccidioidomycosis is self-limiting, and severe initial symptoms have responded to non-specific treatment within a relatively short time. Bedrest for several months was usually prescribed until the patient became asymptomatic and the complement fixation titer of his blood serum subsided or reverted to negative. Another form of therapy has been surgical extirpation of the pulmonary residua of infection to prevent dissemination of the disease. Surgical therapy has been resorted to in the presence of hemoptysis and expanding cavitary lesions to avoid their rupture, or the formation of bronchopleural and pleurocutaneous fistulas, pneumothorax or empyema; solid "coin" lesions have been surgically removed for diagnostic and therapeutic reasons [*31*]. Specific chemotherapeutic treatment for disseminated coccidioidomycosis, however, has not been available and the prognosis has been poor for patients with this form of the disease. Unsuccessful therapeutic agents which have received clinical trials include heavy metals, ethyl vanillate, gentian violet, sodium caprylate, stilbamidine, 2-hydroxystilbamidine, methyltestosterone, diethylstilbestrol, isonicotinic acid hydrazide, para-aminosalicylic acid, nitrogen mustard, potassium iodide, sulfonamides, the antibiotics, penicillin, streptomycin, oxytetracycline, chlortetracycline, prodigiosin, protoanemonin, fradicin, thiolutin, rimocidin, actidione and nystatin, roentgen therapy, culture filtrates and extracts of C. immitis and immune human serum. The problem of evaluating chemotherapeutic agents for coccidioidomycosis and other systemic fungus diseases has been rendered even more difficult by the relative rarity and sporadic distribution of advanced cases of mycoses, the chronicity and pathological nature of the infections, and the prolonged hospitalization and subsequent follow-up usually required of the patient.

AMPHOTERICIN B

In the search for clinically effective antifungal agents, attention is at present focused on amphotericin B,* a polyene, antifungal antibiotic isolated in 1955 by Gold, Stout, Pagano and Donovick [*48*]. This antibiotic is elaborated by a hitherto unnamed species of Streptomyces, isolated from a soil sample taken at Temladora, near the Orinoco River in Venezuela.

Physical, Chemical and Biological Properties. The crystalline, yellow product extracted from both the filtered mycelial mat and the whole culture broth is comprised of two active, antifungal principles, amphotericin A and B, the latter compound being more active against yeasts and yeast-like fungi. Purification and characterization of the amphotericins were accomplished by Vandeputte, Wachtel and Stiller [*49*], who noted that the amphotericins differed from each other in specific rotation, in solubility, and in their ultraviolet absorption maximums and extinction coefficients. These and other properties also made it possible to differentiate the amphotericins from other antifungal antibiotics such as nystatin, rimocidin, trichomycin, ascosin and candicidin [*50*].

Amphotericin A and B have no defined melting points and begin to char at 155° and 170°c., respectively. Elemental analyses indicates that both compounds contain carbon, hydrogen and nitrogen in closely similar proportions and do not contain halogen, sulfur, methoxyl or acetyl groups. A basic moiety, which was identified as an aminodesoxyhexose and named *mycosamine*, has been obtained from the hydrolytic cleavage of amphotericin B; and a tentative empirical formula $C_{46}H_{73}NO_{20}$ has been assigned to the whole compound [*51*]. The amphoteric nature of amphotericin A and B is indicated by their behavior on titration and by their greater solubility in water and aqueous alcohols at high and low pH. Upon addition of mineral acids, both amphotericins form acid salts which are partly soluble in methanol and dimethylformamide. Amphotericin B is also soluble in dimethyl sulfoxide, water + sodium lauryl sulfate, and acidified pyridine. It is insoluble in water, except to a limited extent, at pH 2 and 11, at which points it rapidly loses potency. It is also insoluble in acetone, benzene, aliphatic aldehydes, aliphatic esters, chlorinated hydrocarbons, ethers and other organic solvents. Although it is stable in the dry solid form when kept for one year at 5°c., amphotericin B deteriorates in solution because of hydrolysis and oxidation. Prepared glucose solutions of the antibiotic for intravenous therapy should be discarded after twenty-four hours.

The first intravenous preparation employed in Case 1 produced an unesthetic-appearing, yellow, colloidal suspension when mixed with 5 per cent glucose in distilled water. In subsequent cases, a newer and similarly active water soluble, sodium desoxycholate salt of amphotericin B, which forms a clear, yellow solution, was esthetically more satisfactory. The use of saline solution as a diluent is discouraged by the manufacturer because of its precipitating action on the antibiotic [*54*]. Due either to its large size or its colloidal nature, amphotericin B in aqueous solution is retained by Seitz and ultra-fine glass bacterial filters.

In vitro Spectrum and Mode of Action. In agar dilution tests, amphotericin B exhibits marked fungistatic action against numerous species of non-pathogenic and pathogenic fungi, including Candida albicans, Cryptococcus neoformans, Histoplasma capsulatum, Blastomyces dermatitidis, Blastomyces brasiliensis and Sporotrichum schenckii [*48*]. Other fungus pathogens such as Nocardia asteroides, Aspergillus fumigatus, Phialophora verrucosa, Hormodendrum compactum and Hormodendrum pedrosoi are relatively more resistant to the antibiotic. Amphotericin B was found to be several times more active *in vitro* against yeasts and yeast-like fungi than amphotericin A. It has also been found to be fungicidal for C. albicans at twice the minimal inhibitory concentration [*50*]. Littman and co-workers [*52*] reported the fungistatic level of amphotericin B for this organism to be 0.2 μg./ml. when incubated for forty-eight hours in glucose Penassay broth, and the fungicidal level to be 0.4 μg./ml. Neither amphotericin A nor B is active against bacteria at 50 μg./ml. [*48*].

Acute and Chronic Animal Toxicity [*50,53*]. The mean lethal doses (LD_{50}) of two lots of amphotericin B for white Swiss mice, when injected intraperitoneally, were 2,940 and 5,490 mg./kg., respectively. Both lots of amphotericin B were non-lethal for mice at 8,000 mg./kg. when given orally. Gross pathological examination of mice that survived intraperitoneal injection of the antibiotic revealed the presence of peritoneal adhesions and, in a few instances, yellowish deposits on the surfaces of the liver and spleen. Pale kidneys were noted in mice that had received higher intraperitoneal doses [*50*]. The acute toxicity after intravenous injection in mice was of a higher order than that by other routes of administration, the LD_{50} being 8.7 to 9.4 mg./kg. [*54*]. The toxicity of amphotericin B after intravenous injection varied in different animal species. The majority of deaths in mice occurred within a nine-minute period from the time they received lethal intravenous doses of amphotericin B. Deaths were characterized by ataxia, spasms and terminal collapse [*50,53*]. In cats, a single intravenous infusion of 4.5 to 6.8 mg./kg. amphotericin B within a thirty-minute period produced depression, ataxia, and pulmonary and renal congestion. Intravenous infusions of amphotericin B at 3.5 to 4.8 mg./kg. in dogs caused ataxia and severe, submucosal intestinal hemorrhages, particularly of the

* The trade name of E. R. Squibb and Sons, Division, Olin Mathieson Chemical Corp., for amphotericin B is fungizone.®

duodenum; repeated daily infusions at 1.6 to 3.5 mg./kg. caused similar gastrointestinal lesions [*54*].

Chronic Toxicity. White mice, averaging 25 to 30 gm. in weight, tolerated intraperitoneal administration of amphotericin B in doses varying from 5 to 100 mg./kg. for approximately two months without apparent toxic effect. [*55*]. Amphotericin B failed to produce significant alterations in the formed elements of the blood, in the blood chemical findings or in liver or kidney function tests when given orally to rats at a dose of 1,000 mg./kg., and to dogs at 500 mg./kg. The electrocardiographic tracings in the dogs were unchanged by this treatment [*53*]. A single intramuscular injection of 12.5 mg. of amphotericin B suspension in rabbits produced no irritation, but the material remained deposited in the tissues seven days afterward [*50*].

Absorption and Excretion in Animals [*53*]. Because of its relative insolubility, amphotericin B is absorbed poorly from the gastrointestinal tract of the dog, and presumably as poorly in other animal species. Fasting dogs, given single oral doses of 500 mg. of the antibiotic, exhibited blood plasma levels of only 0.03 to 0.08 μg./ml. amphotericin B up to eighteen hours after the last dose, when assayed by means of a tube-dilution method with Saccharomyces cerevisiae. No amphotericin B could be detected in the heart, liver, spleen, brain or kidneys of dogs twenty hours after they had received the antibiotic orally in a dose of 500 mg./kg. Following oral administration of amphotericin B to dogs, 65 to 87 per cent of the total daily dose was recovered from the stool during one twenty-four-hour period. Levels of amphotericin B in urine were slightly higher in fed dogs than in fasted dogs. Dogs given single oral doses of 250 and 500 mg. of amphotericin B excreted less than 600 μg. (0.1 per cent of the daily dose) in the urine during a five-hour period after the sixth dose day [*53*]. A considerable amount of the sodium salt of amphotericin was absorbed onto the red blood cells of the dog [*50*]. This amount can be freed by lysis of the erythrocytes with 1 per cent saponin and assayed. The free compound (mixture of amphotericin A and B) was distributed equally between the erythrocytes and the plasma [*50*].

Experimental Animal Infections. Sternberg, Wright and Oura [*55*] observed that, in mice infected intraperitoneally with C. immitis, amphotericin B suspension, in daily doses ranging from 10 to 25 mg./kg., produced a protective and therapeutic effect when administered intraperitoneally twenty-four hours after infection. A protective effect also was noted with oral amphotericin B, although at autopsy nine of the ten infected mice were still culturally positive.

Steinberg and co-workers [*56*] reported a prolongation of life of groups of mice that had been infected intravenously with C. albicans, C. neoformans and H. capsulatum and treated subcutaneously with amphotericin B suspension. They also observed that amphotericin B was more effective *in vivo* than the A form, although less soluble. Louria, Feder and Emmons [*57*] noted that a highly protective effect was produced in mice that had been infected intravenously with C. neoformans and H. capsulatum and treated with amphotericin B suspension, either intraperitoneally or orally, in daily dosages of 15 to 150 mg./kg. When a daily intraperitoneal dose of 50 mg./kg. amphotericin B was administered one day after infection and daily for eleven days, 45 per cent of Histoplasma-infected animals were still culturally positive at the conclusion of treatment. Baum, Rubel and Schwarz [*58*] reported increased survival of hamsters that had been infected intraperitoneally with H. capsulatum, when they were treated subcutaneously with 13.3 mg./kg. amphotericin B suspension daily for twenty-eight days. Eighteen days after discontinuation of therapy, tissues of these animals showed a significant decrease in the number of organisms and less pulmonary involvement than the control animals. At autopsy, large amounts of antibiotic were noted at the injection sites, thus reflecting the relative tissue insolubility of amphotericin B.

Therapeutic Trial in Man. Fiese [*59*] obtained a beneficial clinical effect with oral amphotericin B in a twenty-seven year old white man with generalized coccidioidomycosis. The patient received 2.4 gm. of the drug daily by the oral route for three months, for a total dosage of 200 gm. During this period an extensive granuloma of the face showed signs of healing, as indicated by closing of the sinuses and lessening of edema. There were no toxic side effects of the medication. Blood serum levels were not reported.

Littman [*60*] reported a beneficial clinical effect of intravenous amphotericin B in two patients with acute and chronic coccidioidal osteomyelitis, respectively. Adequate and sustained blood serum levels of amphotericin B, assayed by means of tube-dilution method with C. albicans, were achieved by intravenous administration. A number of toxic side effects of intravenous medication were noted. These were minimized by administering the antibiotic in glucose solution at a slow rate and by giving it on alternate days. Oral amphotericin B in one patient was of no clinical value, however, and failed to produce assayable serum levels.

PROCEDURE FOR BIOASSAY OF AMPHOTERICIN B IN BODY FLUIDS

Technic of bioassay involved the visual estimation of the growth inhibitory effect of patient's serum or body fluid on a standard inoculum of C. albicans,

FIG. 5. Bioassay of amphotericin B in body fluids. Patient's serum, added to the assay medium, has produced a fungistatic effect on the growth of the assay organism, C. albicans.

Squibb No. 1539, contained within a glucose-enriched Penassay broth.*

Preparation of the organism and the inoculum: The assay organism was cultivated for forty-eight hours at 37°C. on yeast extract agar† slant. A smooth saline suspension of the organism, the final turbidity of which measured 50 per cent light transmission with the photoelectric colorimeter (Lumetron Model 401A, red filter, 650 mμ.), served as the inoculum. Stock cultures of the organism were maintained in sterile skimmed milk at −20°C.

The growth characteristics and slow rate of growth of C. immitis in liquid culture medium, as well as the hazards involved in the handling of the culture, rendered it unsuitable for use as an assay organism. An added problem was the deterioration of amphotericin B in solution [*52*]. The rapid growth of C. albicans and the production of uniform turbidity in liquid culture medium rendered this species far more satisfactory for use in the test.

Preparation of bioassay culture medium:

Penassay medium, dehydrated, Difco	13.2 gm.
Dextrose	2.5 gm.
Distilled water	250 ml.

The ingredients were dissolved in distilled water in a 500 ml. Erlenmeyer flask, autoclaved at 121°C. (15 pounds) for fifteen minutes and cooled to 20°C., after which 10,000 units of penicillin and 25,000 μg. of streptomycin were aseptically added to provide concentrations of 40 units and 100 μg./ml., respectively. The bioassay culture medium was mixed thoroughly with the antibiotics and stored in the refrigerator at 5°C.

* Beef extract 0.15 per cent; powdered yeast extract 0.15 per cent; peptone 0.5 per cent; dextrose 0.1 per cent; NaCl 0.35 per cent; K_2HPO_4 0.368 per cent; KH_2PO_4 0.132 per cent; pH 7.0.

† Powdered yeast extract 0.3 per cent; peptone 0.5 per cent; dextrose 1 per cent; agar 2 per cent; pH 5-6.

Test procedure: On the day of determination, 0.5 ml. amounts of this culture medium were aseptically dispensed into sterile Wassermann tubes, eight tubes being required for each bioassay. (Table 1.) Body fluids under test were then added in decreasing quantities to each of the eight tubes, and the deficiencies in total volume in each tube were made up with sterile saline solution. One drop (0.07 ml.) of saline suspension of C. albicans cells served as the inoculum, which was delivered with a sterile 1 ml. pipette. This inoculum contained approximately 1 by 10^5 yeast cells which did not impart visible turbidity to the culture medium. Tubes were incubated for twenty-four hours at 37°C., then shaken and examined visually with an ordinary source of illumination for the presence or absence of turbidity (growth). Those tubes which contained fungistatic dilutions of the test fluid remained clear after twenty-four hours of incubation, while the remaining tubes were turbid. (Fig. 5.) Blood serum obtained before the start of amphotericin B infusion was assayed simultaneously with the test serum. Although the use of saline solution in the test is theoretically objectionable because of its action on amphotericin B, bioassays were unaffected by its use.

TABLE 1
TUBE ARRANGEMENT FOR BIOASSAY OF AMPHOTERICIN B IN BODY FLUIDS

Ingredients	Tube Number							
	1	2	3	4	5	6	7	8
Sterile stock assay medium (ml.)	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Blood serum, spinal fluid, etc.	0.5	0.3	0.2	0.1	0.05	0.03	0.02	0
Sterile saline (ml.)	0	0.2	0.3	0.4	0.45	0.47	0.48	0.5
Suspension of C. albicans (drop)	1	1	1	1	1	1	1	1
Level of Amphotericin B μg./ml.*	Turbidity Readings, 24 hr. Incubation at 37°C.							
0	+	+	+	+	+	+	+	+
0.18	−	+	+	+	+	+	+	+
0.30	−	−	+	+	+	+	+	+
0.45	−	−	−	+	+	+	+	+
0.9	−	−	−	−	+	+	+	+
1.8	−	−	−	−	−	+	+	+
3.0	−	−	−	−	−	−	+	+
4.5	−	−	−	−	−	−	−	+

* Computed on basis that the 24 hour MIC of amphotericin B for C. albicans is 0.09 μg./ml.

In order to compute the amphotericin B level of the test fluid by the technic described, it was necessary to ascertain the minimal inhibitory concentration (MIC) of amphotericin B for C. albicans, Squibb No. 1539, under approximately the same culture conditions. The medium used for this purpose was single strength, 1 per cent glucose Penassay broth containing penicillin and streptomycin, to which 10 per cent sterile serum (horse, rabbit or human) had been added. Sufficient quantities of amphotericin B solutions in 100 and 1000 μg./ml. concentrations, dissolved in filter-sterilized, dimethyl sulfoxide (Stefan Chemical Co., Chicago, Ill.), were aseptically added to 10 ml. quantities of glucose Penassay broth to provide stock antibiotic concentrations varying from 0 to 3.0 μg./ml. After thorough mixing, 1 ml. aliquot of each concentration was aseptically transferred to a series of sterile Wassermann tubes, each of which then received one drop (0.07 ml.) of C. albicans suspension. After incubation at 37°C. for twenty-four hours, the tubes were examined for the presence or absence of turbidity. Performed by this method, MIC values of amphotericin B for C. albicans, Squibb No. 1539, averaged 0.09 μg./ml. at twenty-four hours. Solutions of amphotericin B were freshly prepared each week with dry, standard antibiotic (E. R. Squibb & Sons) that was continuously maintained in a desiccator in the dark at 5°C. Dimethyl sulfoxide proved to be neither fungicidal nor fungistatic in the concentrations employed. MIC values were redetermined at weekly intervals.

Body fluids were assayed unfiltered, since both suspension and soluble forms of amphotericin B were retained by Seitz and pyrex glass bacterial filters. Assay of hemolyzed blood serums was avoided. Unhemolyzed specimens were assayed on the day of collection, although storage of serums in the frozen state at − 20°C. did not appear to affect the bioassay. Since urea in normal urine may exert a growth inhibitory effect on C. albicans, bioassay of urine for amphotericin B is inaccurate.

Computation of serum level:

$$\text{Amphotericin B level in test fluid, } \mu\text{g./ml.} = \frac{1}{\text{smallest volume of test fluid showing fungistatic effect}} \times \text{MIC of amphotericin B for C. albicans, No. 1539, } \mu\text{g./ml.}$$

If an end point of fungistasis was not reached, the test was repeated with 1:10 saline dilution of the test fluid.

In Vitro Sensitivity of C. Immitis to Amphotericin B. The minimal inhibitory concentration (MIC) of amphotericin B for eight human isolates of C. immitis was 0.5 μg./ml. after four days incubation at 37°C. (Table II.) The inoculum for each tube consisted of one drop (0.07 ml.) of a ground, mycelial and arthrospore saline suspension which was prepared from nine-day-old growth on slants of Sabouraud dextrose agar enriched with biotin and thiamine. The high in vitro susceptibility of C. immitis to amphotericin B was not affected by the presence of 10 per cent horse serum in the culture medium. Higher MIC values obtained at seven and eleven days incubation were believed to be due to decomposition of the antibiotic.

TABLE II
MIC OF AMPHOTERICIN B FOR COCCIDIOIDES IMMITIS IN 1 PER CENT GLUCOSE PENASSAY BROTH, WITH AND WITHOUT HORSE SERUM (μG./ML.)

Organisms	Days Incubation at 37°C.					
	Without Serum			With 10% Horse Serum		
	4	7	11	4	7	11
C. immitis 860	0.5	0.5	0.5	...	...	...
C. immitis 861	0.5	0.5	0.5	...	...	...
C. immitis 862	0.5	0.7	1.0	...	...	...
C. immitis B. T. (Case IV)	0.5	0.7	1.5	...	...	...
C. immitis P. W. (Case II)	0.5	0.5	0.7	0.5	0.5	0.5
C. immitis P. H.	0.5	0.7	1.5	...	...	...
C. immitis W. D. (Case I)	0.5	0.7	1.0	0.5	0.7	1.0
C. immitis G. R. (Case III)	0.5	0.7	1.0	0.5	0.5	0.5

This had been previously noted by Littman, Pisano and Lancaster [*52*], who reported that the MIC of amphotericin B for C. albicans increased from 0.09 μg./ml. in twenty-four hours to 5 μg./ml. in seven days.

CASE REPORTS

Four cases of coccidioidomycosis are presented, two in detail.

CASE I. A twenty-nine year old white male chemist entered the Mount Sinai Hospital, New York, for the first time on August 27, 1956 with complaints of an enlarged, tender, right knee which had been draining for approximately seventeen years.

The history revealed that at three years of age, in 1930, right mastoiditis had developed, following which he was hospitalized for a widely disseminated staphylococcal osteomyelitis. In the ensuing six years of hospitalization, which was in the pre-chemotherapeutic era, treatment consisted of incision and drainage, and immobilization of the affected extremities. He was discharged from the hospital at the age of nine years, at which time the infection had been arrested. For the next two years he resided in Houston, Texas. At the age of eleven, in 1938, for reasons of health, he moved to Scottsdale, Arizona, a dusty, dry

area in the southwest, highly endemic for coccidioidomycosis. During a one-year residence in Scottsdale he made frequent horseback trips into the desert, where he was exposed many times to Coccidioides-infected dust. During this period, however, he had no bouts of fever, respiratory infections, arthralgias or skin rashes attributable to coccidioidomycosis. In 1939, three months after moving to California, an abscess of the right knee developed which, in retrospect, undoubtedly was a metastatic coccidioidal lesion. Because of the mistaken impression that this represented a recrudescence of the previously existing staphylococcal osteomyelitis, he was returned to the East, where he was hospitalized for fifteen months. The lesion was incised and drained, and finally healed. From 1940 until the present time, the patient did not visit any areas endemic for coccidioidomycosis.

In 1948, following an asymptomatic period of eight years, the patient, then twenty-one years of age, again had pain and edema of the right knee. By 1950, monthly aspiration of the knee joint was necessary. It usually yielded approximately 200 ml. of cloudy, yellowish, synovial fluid. A diagnosis of chronic synovitis was made, and an arthrotomy of the knee joint was performed in June, 1952. At operation the undersurface of the patella appeared softened and was shaved, intra-articular adhesions were excised, and a loose medial cartilage was removed. The synovial membrane was found to be hypertrophic and erythematous. Pathological examination of the membrane revealed edema, infiltration with chronic inflammatory cells, and a few tubercles containing foreign body giant cells, but neither bacterial nor fungal parasites were observed. A flexion contraction of the leg which resulted was corrected, following which a draining sinus of the knee appeared. In the next year the patient fell on his right knee and his extremity was immobilized with a plaster cast for one month. His knee continued to cause pain and discomfort, however, until 1956. At that time a second operation was performed and a granular mass was removed from the right popliteal region, after which another draining sinus developed. A clinical diagnosis of tuberculosis of the right knee was made without a positive culture for Mycobacterium tuberculosis or a positive biopsy for acid-fast bacilli. The patient received a course of streptomycin which had no apparent effect. A diagnosis of coccidioidomycosis was next entertained, but a coccidioidin skin test was negative and C. immitis could not be isolated from the draining sinus. Amputation of the extremity was recommended by several consultant orthopedic surgeons but was refused. In July, 1956, a third operation was performed on the knee and an abscess involving the entire upper medullary cavity of the right tibia was discovered. The abscess was filled with soft, pliable tissue without pus, and communicated posteriorly with the sinus in the popliteal fossa. Profuse bleeding occurred during removal of the tissue.

Examination of the stained tissue sections revealed a few poorly defined coccidioidal spherules without endospores. Fungus cultures of excised bone marrow and abscess material on liver-spleen glucose blood agar [*61*] and Littman oxgall agar [*62*] yielded many colonies of C. immitis. Bacterial cultures yielded an antibiotic-resistant, hemolytic Micrococcus pyogenes var. aureus. Skin tests with coccidioidin (1:100) were 2 +, histoplasmin (1:1000) 3 +, blastomycin (1:1000) negative, torulin (1:1000) negative, and tuberculin, first strength (PPD) 1 +. Serological studies revealed a positive complement fixation titer of 1:32 for coccidioidomycosis and a negative precipitin titer. A diagnosis of chronic coccidioidal osteomyelitis and staphylococcal osteomyelitis was made and the patient was admitted to the hospital for treatment with amphotericin B.

Physical examination revealed a 126 pound, chronically ill, white man who walked with the aid of crutches and who appeared older than his chronological age of twenty-nine years. He had many deep, healed osteomyelitis scars on the left shoulder, orbital region, ankle, right knee and right foot, as well as a flexion contraction of the right arm. Other physical findings were deafness of the right ear, a blood pressure of 150/100 mm. Hg, enlarged deep right inguinal lymph nodes, and an enlarged, tender right knee. A healed scar measuring 15 cm. long was noted on the anteromedial aspect of the upper half of the right leg. The upper half of the scar contained a draining sinus, bordered by numerous excrescences of reddened, firm, granulation tissue from which a copious, serosanguineous fluid exuded. (Fig. 6.) A second smaller draining sinus, 2 cm. in diameter, also was present in the right popliteal fossa. The range of motion of the right extremity was limited to an arc of 90° to 175°. The heart, lungs and abdomen were normal.

Laboratory studies revealed a hemoglobin of 12.5 gm. per cent; red blood cell count, 4,200,000/cu. mm.; white blood cell count, 8600/cu. mm.; differential cell count: polymorphonuclears, 57 per cent; band cells, 5 per cent; lymphocytes, 28 per cent; monocytes, 7 per cent; eosinophils, 3 per cent; blood platelets, 282,000/cu. mm.; erythrocyte sedimentation rate, 112 mm./hour (Westergren); blood urea nitrogen, 15 mg. per cent; creatinine, 2.9 mg. per cent; blood sugar, 103 mg. per cent; total serum protein, 6.1 gm. per cent; serum albumin, 2.4 gm. per cent; serum globulins, 3.2 gm. per cent; and a 2 + proteinuria. The serum electrophoretic pattern revealed a shift in globulin distribution with a preponderance of alpha$_2$ and beta globulins and lowered gamma globulin.

Roentgenograms revealed an extensive destructive lesion involving the right tibial metaphysis, diaphysis and epiphysis, as well as the adjacent subperiosteal bone. (Fig. 7.) There was little surrounding bone reaction. The destructive process was a panosteitis

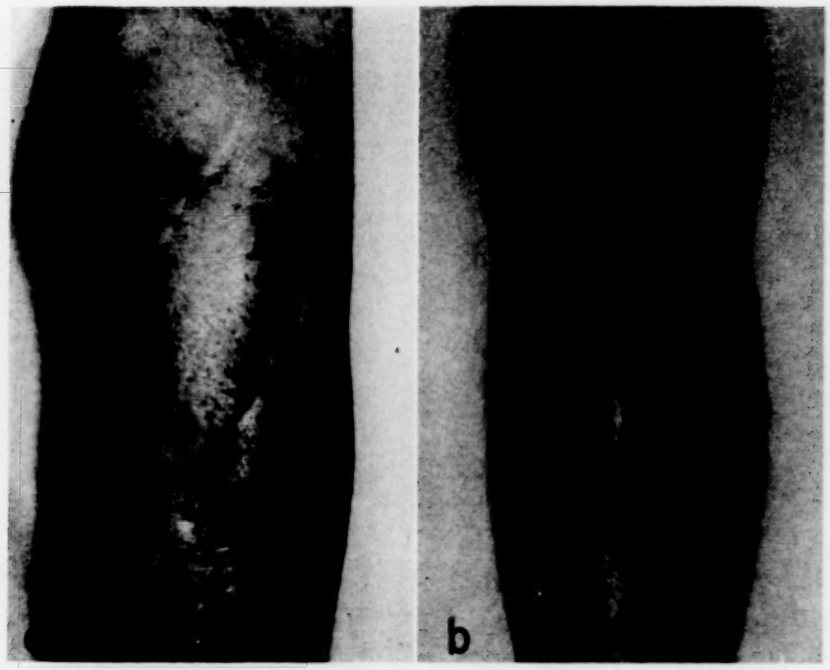

FIG. 6. Case 1. a, Chronic coccidioidal osteomyelitis of the right knee. b, The draining sinuses healed completely coincident with intravenous amphotericin B therapy.

involving the medullary cavity, cortex and periosteum. Under fluoroscopic observation, iodochlorol® (Searle) was slowly injected into the draining mediolateral sinus tract of the knee and roentgenograms were taken. Examination of these films revealed the presence of intercommunicating sinuses extending from the point of instillation posteriorly to the popliteal sinus tract and thence superiorly along the posterior surface of the femur for a distance of approximately 8 cm. (Figs. 7C and D.)

A complete roentgen survey of the bones, including the ribs, failed to reveal any other coccidioidal osseous lesions. Cultures of blood, urine, sputum and sternal bone marrow were negative for C. immitis. Histological examination of sternal bone marrow was negative, except for an increase in eosinophilic cells. Renal biopsy revealed extensive amyloid deposition, focal areas of tubular damage with cloudy, swelling, hyaline droplet changes and epithelial regeneration, all of which were attributed to the chronic osteomyelitis. Electrocardiograms revealed only minor changes.

Tablets of amphotericin B (Lot No. 691-11/15-A-5) were administered orally in divided doses, in amounts increasing gradually from 3 gm. per day until a daily dose of 7.2 gm. was reached on the fourteenth day of treatment. This dosage was maintained for twenty-four days more without signs of gastrointestinal irritation or other side effects, until a total of 226 gm. had been administered. Significant changes in blood chemical findings did not occur during this time. In addition, the patient also received daily infusions of 15 to 30 million units of penicillin for treatment of the coexisting staphylococcal osteomyelitis. No significant progress was made during therapy with oral amphotericin B and intravenous penicillin. In fact, pain, tenderness and edema of the knee increased and the draining sinuses appeared unchanged. During the period of oral therapy, numerous serum amphotericin B assays failed to reveal any drug in the blood serum. A single oral dose of a mixture of 2 gm. of amphotericins A and B (Lot No. 691-11/15-J) failed to produce either fungistatic or fungicidal effect of the patient's blood serum, drawn one and one quarter, three and one-quarter and five and one-half hours after administration and measured by the aforementioned bioassay technic. It was concluded from these observations that the oral preparations of amphotericin A and B employed were too poorly absorbed from the gastrointestinal tract to be clinically and systemically effective, and they were discontinued.

On October 7, 1956, a suspension of amphotericin B for intravenous use (Lot No. 691-14/15-4-F) was made available and was administered to the patient. Coincident with the start of intravenous amphotericin B therapy and during the ensuing four-month period of treatment the patient showed progressive improvement. Tenderness, pain and edema of the knee subsided shortly after the start of intravenous amphotericin B, and complete healing of the lesions eventually followed. (Fig. 6b.) The first intravenous dose of amphotericin B, 50 mg. (0.8 mg./kg.), was suspended in 500 ml. 5 per cent glucose in distilled water, and was administered over an eight-hour period. This was fairly well tolerated. Since the manufacturer did not recommend normal saline solution as a diluent for

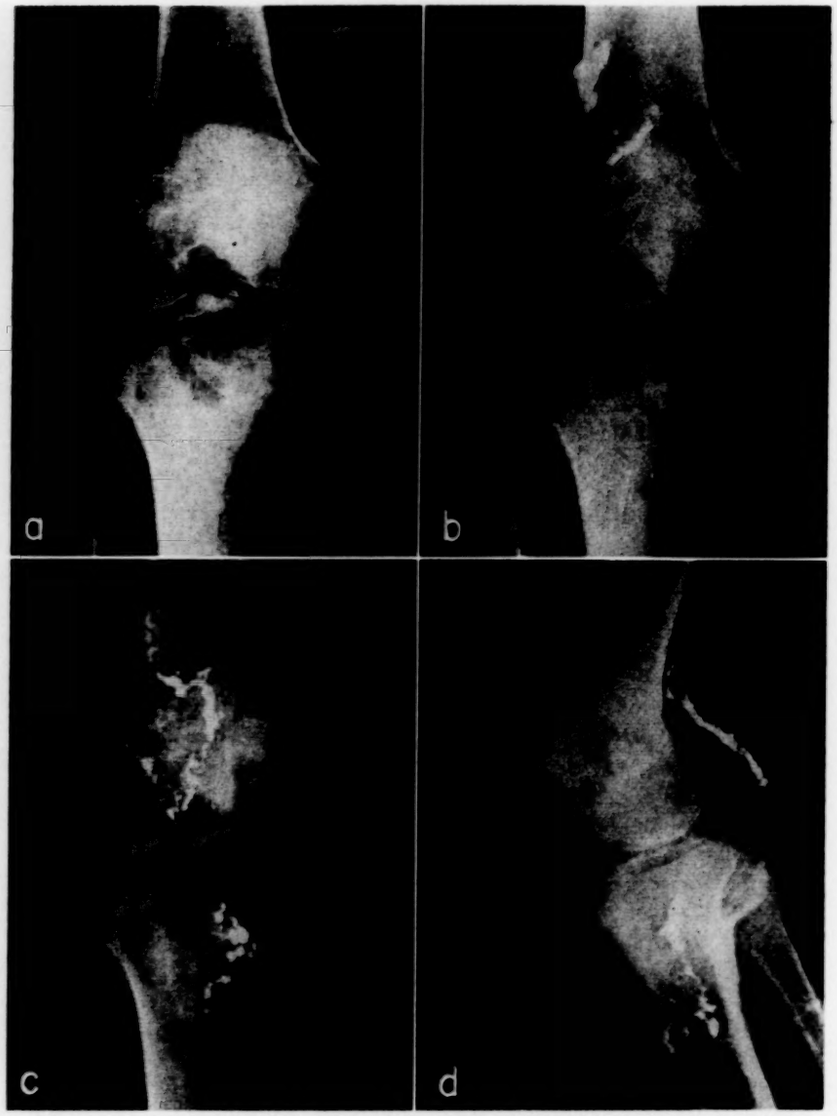

FIG. 7. Case 1. Chronic coccidioidal osteomyelitis of the right knee. (*a*) Destructive lesion of the right tibia involving medullary cavity, cortex and subperiosteum. There is little surrounding bone reaction. (*b*) Same lesion seven months later, two months after cessation of intravenous amphotericin B therapy. Previously destroyed areas show evidence of repair and subsidence of subperiosteal reaction. (*c* and *d*) Iodochlorol injection of the mediolateral sinus tract before intravenous amphotericin B therapy (see Fig. 6) revealed the presence of intercommunicating sinuses extending to the popliteal region and superiorly along the posterior surface of the femur.

intravenous amphotericin B, it was not employed at any time. The daily dose of amphotericin B in glucose solution was gradually increased until, at the close of a three-week period, it had reached 150 mg. (2.6 mg./kg.). At this time the patient's blood urea nitrogen rose from a normal of 15 to 50 mg. per cent, which necessitated discontinuation of the antibiotic. The antibiotic was then withheld for three weeks until the blood urea nitrogen level returned to normal. Amphotericin B was started again and administered on alternate days. Doses did not exceed 100 mg. (1.6 mg./kg.). Azotemia did not recur and thirty-three intravenous doses were consequently administered without complication. Cultures of the sinus tracts became negative for C. immitis shortly after the start of intravenous therapy. Some side effects of amphotericin B, such as nausea, vomiting, flushing, perspiration, fatigue and drowsiness, were noted during infusions. Chilliness and febrile reactions also occurred, but these could be controlled with salicylates and, although less so, with antihistamine drugs. Transitory episodes of vestibular disturbances were encountered on several occasions. The side reactions to intravenous amphotericin B were temporary in nature, however, and subsided when the medication was discontinued. The serum electro-

phoretic pattern became normal after the twenty-first infusion of amphotericin B.

During the months of December, 1956 and January, 1957, 100 mg. of amphotericin B in glucose solution were administered every other day. There were no significant alterations in the patient's hemoglobin, white blood cell count, blood urea nitrogen or liver function tests. The serum electrolytes remained normal and no changes were observed in electrocardiograms. The incidence of drug-induced thrombophlebitis with amphotericin B was no greater than that observed with other infusion medications such as penicillin. A total of fifty-six intravenous infusions of amphotericin B, totalling 4,700 mg., was administered over a period of 118 days.

Doses of 25 and 50 mg. of amphotericin B (Lot No. 691-14/15-4-G) (0.4 to 0.8 mg./kg.), suspended in 500 ml. 5 per cent glucose in distilled water and administered intravenously over a seven-hour period, failed to produce a measurable blood serum level by the aforementioned assay technic. (Table III.) Doses of 100 and 140 mg. (1.6 to 2.4 mg./kg.) given in the same manner, however, produced blood serum levels of 0.9 and 1.8 μg./ml., respectively. The persistence of amphotericin B levels in the blood serum twelve and eighteen hours after the antibiotic had been discontinued suggested a high renal threshold of the drug, and that conjugation by the liver was not rapid.

Additional therapeutic measures consisted of (1) immobilization of the extremity in a half-spica plaster cast, with a window over the lesion; (2) intravenous penicillin in doses varying from 15 to 30 million units daily for treatment of the coexisting staphylococcal osteomyelitis; (3) injections of autogenous staphylococcal, heat-killed, vaccine; (4) frequent injections of vitamin B complex. The MIC of penicillin required to inhibit M. pyogenes var. aureus increased from 0.5 units per ml. in September, 1956, to 2.0 units per ml. in January, 1957. During one infusion of 15 million units of penicillin in 500 ml. 5 per cent glucose in distilled water, the patient's blood serum assayed 33 units penicillin per ml.

Serologic studies at the end of therapy revealed a decrease to 1:16 in the complement fixation titer for coccidioidomycosis and also showed a negative precipitin titer, findings which were associated with clinical improvement. At this time skin tests with coccidioidin (1:100) were 1 +, histoplasmin (1:1000) 2 +, blastomycin (1:1000) negative, torulin (1:1000) negative, and tuberculin, first strength (PPD) 1+.

Roentgenograms of the right knee in January, 1957, revealed that previously destroyed areas of the tibia showed evidence of repair and subsidence of subperiosteal reaction. The patient was fitted with a brace to assist in weight-bearing, and on February 15, 1957 he was discharged from the hospital with a healed knee. He was requested to return at periodic intervals for evaluation and when seen six months later he had gained twenty pounds. He had remained asymptomatic during this time and his right knee and foot were still healed. The complement fixation titer of the serum had not changed significantly in five months and was still 1:16. The precipitin titer remained negative.

CASE II. A twenty-three year old Negro man was admitted to the Veterans Administration Hospital, Bronx, New York, for the first time on December 26, 1956, with complaints of swelling and pain of the left knee and left extremity of approximately three years' duration.

The history revealed that the patient had resided in Mississippi until the age of ten years. In 1944 he moved to Los Angeles, California, where he lived until 1951. Before his induction into the U.S. Army that year he had made frequent trips to Bakersfield, California, one of the most highly endemic areas for coccidioidomycosis in the Southwest. During and after this period, however, he had no episodes of fever, respiratory infections, skin rashes or arthralgias attributable to coccidioidomycosis. While stationed in Korea in 1951, he fell from a military vehicle, injured his left knee, and was hospitalized. He received symptomatic treatment and recovered. In 1953 the same knee became enlarged and painful after a second "injury" and treatment consisted of heat and immobilization. The knee remained somewhat enlarged, however. In 1955 he had increased swelling and tenderness of the left knee. Fluid was aspirated from the joint and hydrocortisone was injected, with consequent reduction of the swelling and pain. At the time of his discharge from the service in June, 1956, nevertheless, the patient still had an enlarged, stiff and painful left knee. In September, 1956, after receiving a third injury in an auto accident, gross enlargement of the knee developed which made walking impossible. A progression of signs and symptoms followed, including weight loss of twenty pounds, night sweats, 4 + pitting edema of the left lower extremity, and the appearance of systolic and diastolic heart murmurs. The patient was hospitalized in December, 1956, with a diagnosis on admission of traumatic arthritis of the knee and subacute bacterial endocarditis.

Physical examination revealed a 140 pound Negro man, with a swollen and edematous left thigh, enlarged lower extremity and a grossly enlarged, acutely tender, warm left knee containing excessive joint fluid. (Fig. 8.) The left extremity was held in flexion, as movement caused considerable pain. Point tenderness was absent in the remainder of the extremity, however, and there was no evidence of superficial thrombophlebitis. Homans' sign was absent. The deep inguinal lymph nodes on the left were considerably enlarged. Two splinter hemorrhages were observed in the nail beds of the right index and middle fingers. The blood pressure was 120/65 mm. Hg, the pulse rate was 88 per minute. Auscultatory examina-

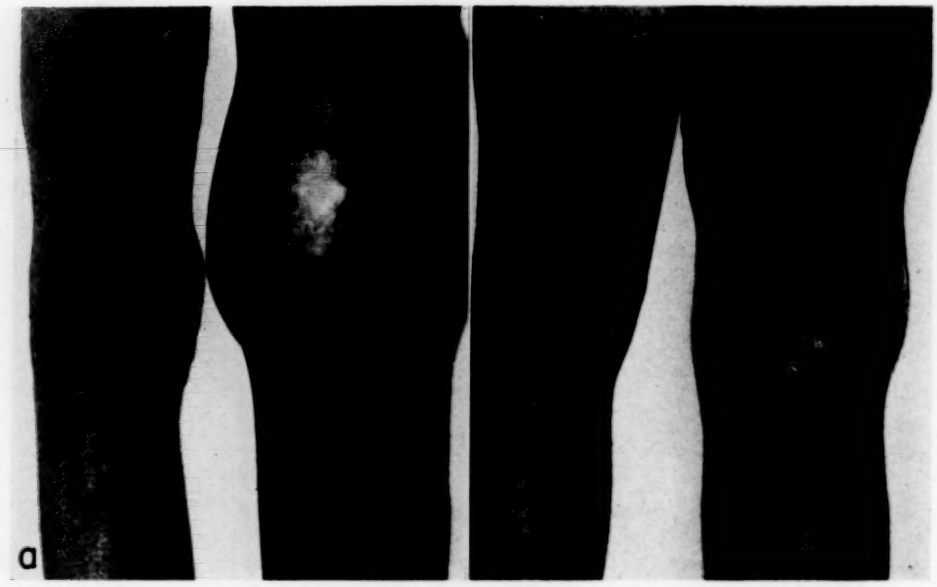

FIG. 8. Case II. Coccidioidomycosis of the knee. (*a*) Before therapy, the grossly enlarged left knee contained excessive joint fluid from which C. immitis was recovered. (*b*) Marked clinical improvement was noted after five months of intermittent therapy with intravenous amphotericin B, combined with surgical drainage and other supportive measures.

tion of the heart revealed a short, grade 2, loud, harsh, apical systolic murmur and a loud, grade 3, decrescendo diastolic murmur at the base, but loudest at Erb's point, with radiation of the murmur to the aortic area and downwards towards the epigastrium. The liver and spleen were not enlarged. Roentgenogram of the left knee revealed pericapsular swelling, synovitis and minimal destructive changes consistent with either tuberculous or pyogenic osteomyelitis.

Laboratory studies revealed a hemoglobin of 11.8 gm. per cent; red blood cell count, 4,000,000/cu. mm.; hematocrit, 39 per cent; white blood cell count, 7,200/cu. mm.; differential cell count: polymorphonuclears, 58 per cent; lymphocytes, 24 per cent; monocytes, 6 per cent; eosinophils, 12 per cent; blood platelets, 198,000/cu. mm.; erythrocyte sedimentation rate, 70 mm./hour (Westergren); blood urea nitrogen, 6 mg per cent; serum bilirubin, 0.3 mg. per cent; total serum protein, 7.7 gm. per cent; serum albumin, 3.0 gm. per cent; serum globulins, 4.7 gm. per cent; cephalin flocculation test, 2 +; alkaline phosphatase, 4.2 units (Shinowara); thymol turbidity, 5.7 units. The serum electrophoretic pattern showed an increase in alpha$_2$ and gamma globulin components. Urinalysis on the day following admission revealed acid reaction; specific gravity, 1.030; albumin, trace; sugar, negative; 50 white blood cells per high power field. Electrocardiogram revealed elevation of ST segments in the standard and precordial leads and T wave changes in leads V_3 to V_6. Chest x-ray was normal.

The first diagnostic impressions were of deep thrombophlebitis of the left leg or chronic traumatic arthritis of the left knee. The subsequent findings of cardiac murmurs and splinter hemorrhages suggested a diagnosis of subacute bacterial endocarditis. Upon admission of the patient his left knee joint was aspirated, but only 3 ml. of yellow fluid were obtained. This was examined by direct smear and culture and found to be bacteria-free. Fungus disease was not suspected and cultures for fungi were not performed. Although afebrile on admission, the patient became acutely febrile on the fourth hospital day. (Fig. 9.) Despite reports of four consecutive negative blood cultures, treatment for subacute bacterial endocarditis was started with intravenous penicillin (8 million units daily) and intramuscular streptomycin and dihydrostreptomycin (1 gm. of each daily). By the twelfth hospital day there was still no therapeutic response to these antibiotics and a new splinter hemorrhage appeared in the nail bed of the right fourth finger. One week later the patient's hemoglobin had fallen to 10.3 gm. per cent and his temperature reached 102° and 103°F. The daily dosage of intravenous penicillin was increased to 40 million units and erythromycin was added in doses of 2 gm. per day. There were no signs of clinical improvement, however, and the patient began to experience daily temperature peaks of 104° and 105°F. The left knee remained hot, swollen and exquisitely tender, despite the antibiotic therapy. Thirteen blood cultures taken during the first month of hospitalization were negative. On the twenty-seventh hospital day the patient was moved to the operating room where, under anesthesia, a synovial biopsy was taken and 40 ml. of bloody, purulent fluid were aspirated from the left suprapatellar bursa. Gram stain of the fluid revealed a preponderance of polymorphonuclear cells but no bacteria. The biopsied tissue, however, revealed numerous coccidioidal spherules containing endo-

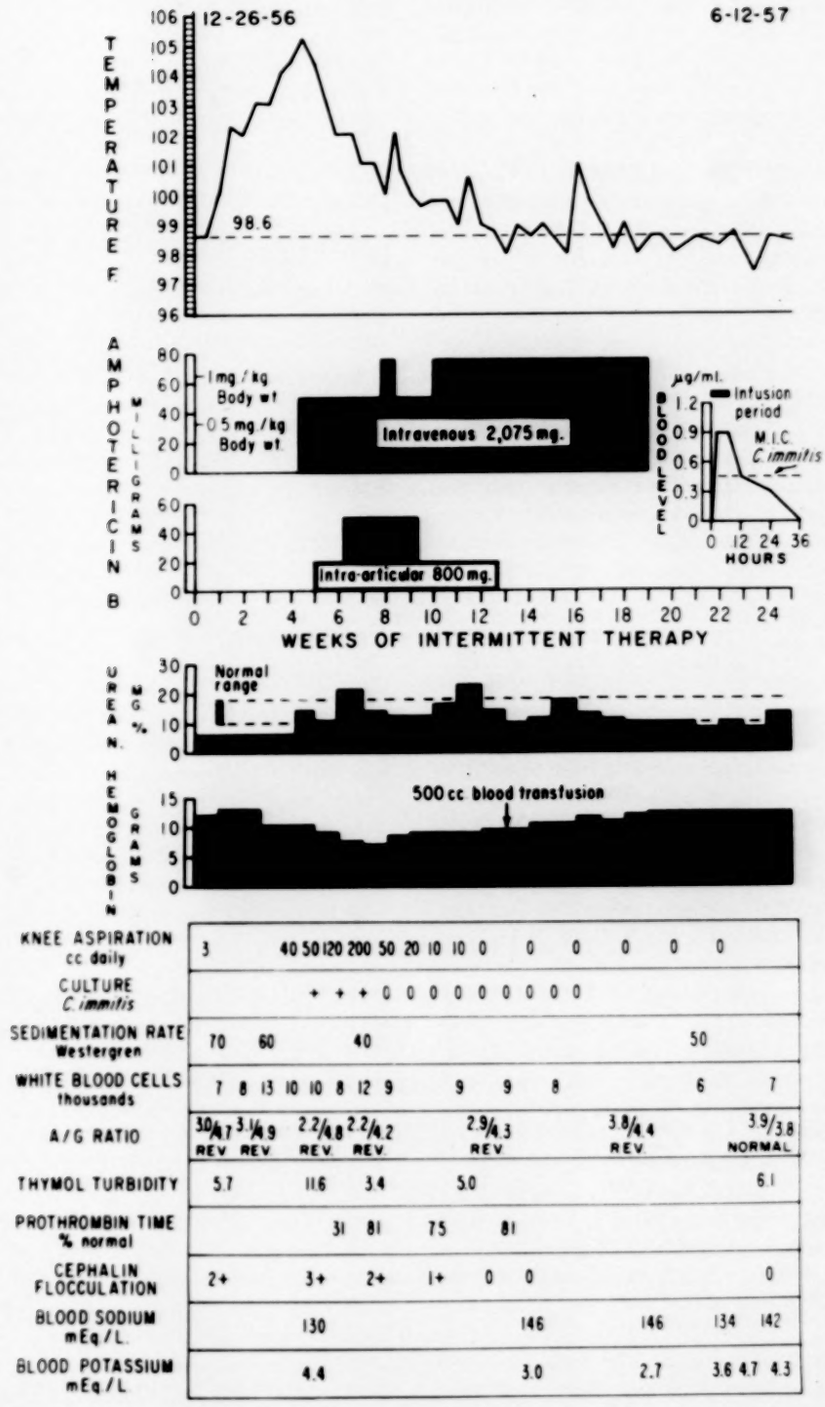

FIG. 9. Case II. Chart showing clinical response to intravenous amphotericin B of a twenty-three year old, 140 pound Negro man with coccidioidomycosis of the left knee.

spores, finally solving this difficult clinical diagnostic problem. Cultures of the knee fluid on Littman's liver-spleen glucose blood agar [*61*], Littman oxgall agar [*62*] and other glucose blood agar culture media yielded many colonies of C. immitis. Cultures of blood, bone marrow, spinal fluid, urine and sputum, however, were all negative for this organism. Serological studies revealed a high complement fixation titer for coccidioidomycosis (1:512) and a negative precipitin titer, indicative of dissemination of the disease as well as a poor prognosis. Skin tests with coccidioidin (1:100), as well as with 1:1000 dilutions

of blastomycin, histoplasmin and torulin, were negative. The patient's strain of C. immitis was found to be inhibited by 0.5 μg./ml. amphotericin B. (Table II.)

On the thirtieth hospital day (January 25, 1957) the patient was given his first intravenous dose of 50 mg. amphotericin B suspension in 250 ml. 5 per cent glucose in distilled water (0.8 mg./kg./day) at the rate of 8 to 10 drops per minute. The solution was inadvertently allowed to flow in too rapidly, introducing 28 mg. of amphotericin B intravenously in forty minutes, following which the patient had a shaking chill. Within five minutes generalized convulsions of the grand mal type developed, which terminated spontaneously within thirty seconds. This was followed by apnea and absence of heart sounds. A blow on the chest with the fist caused the heart to beat again with a regular rate of 95 per minute and the patient immediately regained consciousness. Three minutes later a second convulsion occurred, with the same cardiac abnormality. This again responded to a blow on the chest and an electrocardiogram taken five minutes later revealed a regular sinus rhythm and prolongation of the QT interval to 0.40 seconds. The next day a similar dose of amphotericin B was administered intravenously but this time slowly over an eight-hour period. It was well tolerated. For the first time in two weeks of unremitting high fever, the patient's temperature fell to 97°F. On the following day, without intravenous amphotericin B, the temperature again rose to 102.4°F. The third and succeeding 50 mg. dose of amphotericin B, consisting of a newer, water-soluble sodium desoxycholate salt (Lot No. 691-14/15-7) dissolved in 500 ml. 5 per cent glucose in distilled water, was administered by slow intravenous drip over a period of six to eight hours. In each instance, when the first 5 to 15 mg. of amphotericin B had been infused, a typical drug reaction occurred. This consisted of marked anxiety followed quickly by generalized pain, shaking chills and a febrile reaction, after which the temperature slowly fell to normal and remained there for approximately thirty-six hours. This was followed by a slow spontaneous rise in temperature, until the next dose of amphotericin B was administered. Large doses of acetylsalicylic acid did not adequately control the febrile reaction nor did the antihistamine, chlortrimeton maleate.®

Blood serum levels of amphotericin B achieved by intravenous administration of 50 mg. of the antibiotic in 500 ml. 5 per cent glucose in distilled water, at a rate of 20 drops per minute, were slightly below the minimal inhibitory concentration for C. immitis. (Tables II and IV.) Increasing the dose to 75 mg., or 1.2 mg./kg./day, raised the serum level of amphotericin B to 1.8 μg./ml. during the infusion. Twenty-four hours after discontinuation of the infusion, the serum level was 0.9 μg./ml., or approximately twice the MIC of C. immitis. (Tables II and IV.) With each administration of amphotericin B, on alternate days, the patient showed an increased tolerance to the antibiotic. Within twelve hours following each infusion the patient's temperature markedly declined; by the eighth infusion he was afebrile and able to sit up comfortably in bed after nearly one month of high fever and prostration. There was also an increase in appetite and well-being.

The left suprapatellar bursa was aspirated four times a week and 30 to 50 ml. of bloody, purulent fluid were obtained each time. (Fig. 9.) Starting with the fifth week of hospitalization, intra-articular instillations of 25 mg. amphotericin B, with 10,000 units each of streptokinase and streptodornase, were made into the bursa after it had been aspirated. The drug was well tolerated at this site. After several instillations of the drug had been made into the joint, a sharp reduction was noted in the number of viable cells of C. immitis in the aspirated fluid. On the forty-fifth hospital day two draining sinuses spontaneously appeared on the medial and lateral surfaces of the left knee, discharging copious, sanguino-purulent fluid from which C. immitis was cultured. After the tenth intravenous dose of amphotericin B the patient's general condition improved sufficiently to enable him to undergo a lengthier period of general anesthesia. An extensive incision and drainage of the left knee was performed, revealing a large abscess filled with sanguino-purulent material. This was evacuated, the devitalized tissues were excised, the wound was irrigated with amphotericin B solution and catheter drains were inserted. The left extremity was then immobilized with an open, half-spica plaster cast. An increase in blood urea nitrogen did not occur although the patient continued to receive intravenously, on alternate days, 75 mg. of amphotericin B in 5 per cent glucose in distilled water, as well as intra-articular doses of 25 mg. in distilled water three times a week. Cultures of the knee fluid for C. immitis became negative on the fifty-sixth day of hospitalization, or twenty-seven days after start of amphotericin B therapy. Under the combined regimen of (1) surgical débridement and drainage, (2) intravenous administration of amphotericin B, (3) local instillation of amphotericin B with streptokinase and streptodornase, and (4) immobilization of the extremity, the drainage from the involved knee progressively diminished, finally ceasing on the eighty-seventh hospital day, after a total of twenty-four intravenous and twenty-two intra-articular doses of amphotericin B had been administered. The operative wounds healed quickly, the patient became afebrile and remained so for the remainder of his hospitalization.

On the seventy-first hospital day, 75 mg. of amphotericin B were given intravenously every day instead of on alternate days. This was discontinued after four days when the patient complained of severe nausea, vomiting, abdominal pain, dizziness and tingling of

the fingers. His blood urea nitrogen increased from a normal value of 6 to 23 mg. per cent. When the same dosage schedule was reduced to alternate days, the severity of the side reactions to the drug and the azotemia slowly abated. During the ensuing months of April, May and June, 1957, the patient became completely asymptomatic and showed a gain in weight. Amphotericin B was discontinued after the nineteenth week of intermittent therapy, at which time the patient had received thirty-two intravenous doses totalling 2,075 mg., at a rate of 1.2 mg./kg./day; and twenty-three intra-articular doses, or a total of 800 mg. The incidence of thrombophlebitis was no greater than that encountered with other infusions. Serologic studies at the end of therapy revealed a distinct fall, to 1:128, in the complement fixation titer for coccidioidomycosis and a negative precipitin titer. The fall in complement fixation titer was associated with the patient's clinical improvement.

After six months of hospitalization the patient was discharged on June 28, 1957, ambulating on crutches. He returned three weeks later, however, because of the reappearance of three superficial, draining, cutaneous sinuses of the left knee, from which C. immitis was isolated. The periarticular tissues were mildly swollen, hot and tender. The minimal inhibitory concentration of amphotericin B for C. immitis was found to be unchanged from that of the first isolate, i.e., 0.5 μg./ml. This was taken to indicate that the organism had not become resistant to amphotericin B during the previous course of intravenous therapy. The patient was afebrile and his general physical status was unchanged. The sinuses were débrided surgically, drains were inserted, and amphotericin B was instilled locally as well as administered intravenously. A posterior splint was again applied to immobilize the knee joint. An initial intolerance to intravenous amphotericin B, as evidenced by febrile reactions, was again observed. After eight infusions, however, the patient tolerated doses of 75 mg. (1.2 mg./kg.) intravenously, which produced adequate blood levels. Sixteen intravenous doses of amphotericin B with glucose solution were administered over a period of forty-two days, for a total of 1,206 mg. By the twenty-eighth day, drainage from the sinuses ceased, all signs of inflammation subsided, and cultures of the sinus tracts were negative for C. immitis. At the end of therapy the sinuses had filled-in completely and had become re-epithelialized. On September 17, 1957, a low serum potassium level of 2.7 m Eq./L. was noted and oral potassium therapy was instituted. After correction of the hypokalemic state, the patient was discharged and advised to return for follow-up examinations.

In this patient evidence of involvement of several systems was noted, as follows: *Cardiovascular:* Although the initial clinical impression was subacute bacterial endocarditis, a series of blood cultures were negative for bacteria and fungi. After a specific diagnosis of coccidioidomycosis had been made, however, the electrocardiographic abnormalities, i.e., ST segment elevations in standard and precordial leads and T wave changes in V_3 to V_6, were interpreted as probably due to coccidioidal pericarditis. Cardiac enlargement was absent, the aortic insufficiency never became important hemodynamically, significant changes in heart murmurs did not occur, and the patient failed to show any signs of cardiac decompensation. Shortly after amphotericin B was administered intravenously, precordial T waves in serial electrocardiograms showed a progressive change towards normal, and at the conclusion of the series of infusions the electrocardiogram became normal. It was believed that the pericarditis had been caused specifically by coccidioidomycosis, because of the presence of a high complement fixation titer (1:512) and a negative precipitin test, which indicated dissemination of the disease. It was believed also that the patient had had a prior rheumatic involvement of the aortic valve.

Hepatic: The patient's liver did not become enlarged throughout his illness but various tests indicated the presence of abnormalities of liver function. Upon admission the serum globulin was 4.7 and serum albumin was 3.0 gm. per cent. Hyperglobulinemia persisted until the end of therapy. (Fig. 9.) Abnormalities of the cephalin flocculation and thymol turbidity tests were present. Serum electrophoretic patterns obtained early and late in the hospital course revealed a qualitative increase in the $alpha_2$ and gamma globulin fractions. The serum alkaline phosphatase was abnormal, reaching a maximum value of 12.1 Shinowara units (upper limit of normal, 8.6) on the thirty-fifth hospital day and thereafter gradually falling to normal. Coincident with amphotericin B therapy, the serum proteins, cephalin flocculation test, thymol turbidity and alkaline phosphatase tests returned to normal. This was considered to be indirect evidence that the liver had been involved with C. immitis and that the antibiotic therapy was the remedial factor.

Hematopoietic: On admission, the patient's hemoglobin was 11.8 gm. per cent and the hematocrit was 39 per cent. A fall in hemoglobin occurred during the first month of hospitalization, before amphotericin B therapy was instituted. The red blood cells were hypochromic, with anisocytosis and poikilocytosis. Examination of iliac crest bone marrow at this time revealed adequate megakaryocytes, with marked platelet formation and a normal cellularity. There was an increase in plasma cells to 16 per cent which was consistent with chronic infection and hyperglobulinemia. Stool guaiac tests were positive. After nine intravenous doses of amphotericin B had been administered over a period of seventeen days, the patient's hemoglobin fell to 6.8 gm. per cent and one transfusion of 500 ml. whole blood was given. (Fig. 9.) Roentgenographic studies of the upper gastrointestinal tract revealed evidence of duodenitis. Internal hemor-

rhoids were also found. The severe anemia was attributed to disseminated coccidioidomycosis, associated with gastrointestinal bleeding and possibly also to bleeding internal hemorrhoids. The anemia was not attributed to amphotericin B therapy, since tests for increased hemolysis, such as red blood cell fragility, serum bilirubin, Coombs' test and reticulocyte count were normal on several occasions. By the eighty-seventh hospital day, after twenty-four intravenous and twenty-two intra-articular doses of amphotericin B had been administered, iron therapy with feosol® tablets was begun. By the 130th hospital day a gradual increase in hemoglobin to a level of 13 gm. per cent occurred without further blood transfusions. With this rise there also occurred a mild reticulocytosis, the highest level being 2.3 per cent.

Renal: A persistent pyuria and trace of albuminuria were present on admission. The organisms, Aerobacter aerogenes and Streptococcus fecalis, were cultured from the urine but could not be eradicated even by several separate courses of chloramphenicol, gantrisin® and achromycin.® Numerous urine cultures were negative for C. immitis. An intravenous pyelogram was performed one month after admission, revealing blunting and distortion of the superior calyx of the right kidney, and suggesting chronic pyelonephritis. Although urinary concentration was excellent (specific gravity, 1.030), phenolsulfonphthalein excretion test showed some impairment, 15 per cent phenolsulfonphthalein being excreted in fifteen minutes and 40 per cent in one hour.

After thirty intravenous doses of amphotericin B had been administered, a routine electrocardiogram revealed changes suggestive of hypokalemia, i.e., sinus bradycardia of 60, prolonged QT interval with low T waves in leads 1 and 2, diphasic T waves in leads V_2 to V_6, with prominent U waves in the precordial leads. The serum potassium was found to be decreased from a normal level at admission to 2.8 mEq./L. In view of the patient's normal dietary intake, the source of potassium loss was thought to be in the urine. This was confirmed by the finding of a high twenty-four-hour urinary potassium value of 101 mEq. Despite the hypokalemic state and abnormal electrocardiogram, the patient was asymptomatic. The blood sodium at this time was 146, CO_2 combining power 26.1 mEq./L. Amphotericin B therapy was therefore discontinued. Although supplemental potassium was not given for the next three weeks, the serum potassium rose from 2.8 to 3.6 mEq./L. (Fig. 9.) When oral potassium was then administered, at a rate of 5 gm. daily for the next two weeks, the serum potassium increased to a normal value of 4.7 mEq./L. and the electrocardiograms returned to normal.

Hypokalemia occurred a second time, in the period of relapse in August, 1957, after the patient had received 75 mg. doses of intravenous amphotericin B, on alternate days, for thirteen doses. This was associated with signs of generalized muscular weakness, decreased serum potassium to 2.9 mEq./L. and electrocardiographic changes suggestive of hypokalemia. Antibiotic therapy was stopped and oral potassium administered until the hypokalemic state was reversed.

The following two cases of coccidioidomycosis, to be published in greater detail later, are summarized briefly:

Case III. In a native New Yorker, exposed to Coccidioides-infected dust in California, fever, cough, night sweats and pneumonitis developed, followed by pulmonary and cutaneous lesions, sputum positive for C. immitis, and a complement fixation titer indicative of dissemination of coccidioidomycosis. After a series of intravenous infusions with amphotericin B, the patient became asymptomatic, his sputum and gastric cultures became negative for C. immitis, and his cutaneous lesions healed.

Case IV. In a native New Yorker, exposed in the same manner, a respiratory disease developed characterized by productive cough, sputum and gastric aspirate positive for C. immitis, and pulmonary cavitation. He was treated with a series of intravenous amphotericin B infusions in doses which produced therapeutic serum drug levels, following which he showed clinical improvement. His cough subsided while on therapy, sputum and gastric cultures became negative for C. immitis, and the pulmonary lesions regressed in size. Side reactions to amphotericin B in Cases III and IV consisted of febrile reactions and occasional azotemia which subsided upon discontinuation of the antibiotic.

The changes in serologic reactivity and skin hypersensitivity in three patients are summarized in Table V. A fall in complement fixation titer in Case II and a moderate fall in Cases I and III occurred after therapy with amphotericin B administered intravenously. No significant changes in the degree of skin hypersensitivity to coccidioidin occurred during therapy.

COMMENTS

Clinical experience with oral amphotericin B indicates that it is non-toxic and well tolerated even when administered in large doses. The oral preparation failed to produce assayable blood serum levels on several trials, however, and appeared to be of no clinical value in one patient with coccidioidomycosis. Although oral amphotericin B is poorly absorbed from the gastrointestinal tract, it is highly fungicidal in vitro for C. albicans. The oral antibiotic may be of value, therefore, in the management of gastrointestinal moniliasis, in the treatment of super-

infections of the gastrointestinal tract following the use of broad spectrum antibiotics, and in preoperative, surgical preparation of the patient. Indications of this may be seen in the work of Halde and co-workers [*63*] who noted that oral amphotericin B, administered alone or in combination with tetracycline, caused a marked decrease in the number of yeasts and C. albicans in the stool.

TABLE III

ASSAYABLE LEVELS OF AMPHOTERICIN B* IN BLOOD SERUM COINCIDENT WITH INTRAVENOUS THERAPY (CASE I)

Dose Amphotericin B Suspension* in 500 ml. 5% Glucose in Distilled Water		Duration of Infusion, (hr.)	Time Blood Specimen Obtained	Amphotericin B Blood Serum Level, (μg./ml.)
mg.	mg./kg.			
25	0.4	7	During infusion	0
50	0.8	7	During infusion	0
100	1.6	..	Before infusion	0
		7	During infusion	0.9
		..	12 hr. after discontinuance	0.9
		..	18 hr. after discontinuance	0.9
		..	38 hr. after discontinuance	0
140	2.4	8	During infusion	1.8

* Lot No. 691-14/15-4-G, suspension.

A beneficial clinical effect was noted in four cases of coccidioidomycosis treated with amphotericin B intravenously. The antibiotic is considerably more toxic intravenously than when administered orally. The maximum tolerated intravenous dose of amphotericin B is 1.0 to 1.6 mg./kg./day when administered with glucose solution over a six to eight hour infusion period. An intravenous dose of 28 mg. administered with glucose within forty minutes, however, proved to be almost fatal in Case II. It caused acute prolongation of the QT interval in the electrocardiogram, severe depression of cardiac conduction, and cardiac standstill which, in this instance, was reversed. When administered intravenously at 1.0 to 1.6 mg./kg./day over a six to eight hour period, amphotericin B failed to produce any signs of cardiac toxicity, as evidenced by the absence of abnormalities in the electrocardiogram.

Electrocardiographic abnormalities, presumably due to coccidioidal pericarditis, were noted in Case II before the start of amphotericin B therapy, i.e., ST segment elevations in standard and precordial leads and T wave inversions in leads V_3 to V_6. It is of significance to note that during treatment with amphotericin B intravenously, electrocardiograms became normal. Similar electrocardiographic abnormalities in three patients with pulmonary coccidioidomycosis and pericarditis have been observed by Larson and Scherb [*64*]. Myocardial invasion by C. immitis also appears to be a frequent occurrence, for Forbus and Bestebreurtje [*4*] reported that in fourteen of fifty fatal cases of coccidioidomycosis there were myocardial lesions and Gore and Saphir [*65*] noted eleven instances of myocardial lesions in forty-eight cases. Pericarditis appears to result from the eruption of superficially situated granulomatous lesions located in and just beneath the epicardium [*4*]. In none of the fourteen cases cited was an endocardial lesion found.

TABLE IV

ASSAYABLE LEVELS OF AMPHOTERICIN B* IN BLOOD SERUM COINCIDENT WITH INTRAVENOUS THERAPY (CASE II)

Dose Amphotericin B (Soluble)* in 500 ml. 5% Glucose in Distilled Water		Duration of Infusion (hr.)	Time Blood Specimen Obtained	Amphotericin B Blood Serum Level, (μg./ml.)
mg.	mg./kg.			
50	0.8	6	During infusion	0.45
75	1.2	6	During infusion	1.8
		..	5 hr. after discontinuance	1.8
		..	24 hr. after discontinuance	0.9
		..	48 hr. after discontinuance	0

* Lot No. 691-14/15-7, water soluble sodium desoxycholate salt.

Side effects of intravenous amphotericin B, such as nausea, occasional vomiting, flushing, perspiration, fatigue, drowsiness, chilliness, febrile reactions, anxiety and generalized pain, subsided upon discontinuation of the medication. Febrile reactions to 102°F. occurred frequently, but could be controlled in some patients with the prophylactic use of salicylates or antihistamine drugs. Occasionally it was necessary to interrupt therapy for several hours because of the severity of side reactions. In order to lessen these effects, initial intravenous therapy was started at 0.25 mg./kg. and gradually increased to 1.0 to 1.6 mg./kg. An increased tolerance to intravenous amphotericin B was noted with each succeeding infusion.

The toxicity of intravenous amphotericin B in man appears primarily to involve the kid-

TABLE V

CHANGES IN SEROLOGIC REACTIVITY AND SKIN HYPERSENSITIVITY IN PATIENTS WITH COCCIDIOIDOMYCOSIS, COINCIDENT WITH INTRAVENOUS AMPHOTERICIN B THERAPY

	Case I			Case II			Case III		
	Complement Fixation Titer	Precipitin Titer	Coccidioidin 1:100*	Complement Fixation Titer	Precipitin Titer	Coccidioidin 1:100	Complement Fixation Titer	Precipitin Titer	Coccidioidin 1:100
Before treatment	1:32	Negative	2+	1:256	Negative	Negative	1:32	Negative	1+
During treatment	1:16	Negative	2+	1:512	Negative	Negative	.....	.....	...
After treatment	1:16	Negative	1+	1:128	Negative	Negative	1:16	Negative	...
Follow-up	1:16 (5 mo.)	Negative	...	.....	.....	.....	1:16 (1 mo.)	Negative	...

* Skin reaction to intradermal injection of 0.1 ml.

neys. In almost every instance in which the drug was administered in excessive or too frequent doses, a rising azotemia occurred. Infusions were discontinued when the blood urea nitrogen exceeded 30 mg. per cent, or the non-protein nitrogen exceeded 50 mg. per cent. They were not started again until the azotemia had been alleviated. Azotemia did not occur, however, when intravenous amphotericin B was administered on alternate days in doses of 1.0 to 1.6 mg./kg./day. Adequate blood serum levels of the antibiotic were still present twenty-four hours after an infusion (Tables III and IV), consequently therapy on alternate days was employed routinely. The persistence of amphotericin B serum levels at twelve, eighteen and twenty-four hours after a single infusion of the antibiotic was ascribed to the high renal threshold of the drug, associated with a prolonged biological half-life. A hypokalemic effect was noted in one patient (Case II). This was characterized by high urinary potassium excretion and abnormal electrocardiograms, which slowly returned to normal when the drug was discontinued. Other than this, intravenous amphotericin B appeared to have no effect on urinary concentration or renal excretion.

No deleterious effects on liver function, hematopoietic system or neurologic system could be attributed to intravenous amphotericin B, nor were cutaneous reactions observed. The occurrence of submucosal, intestinal hemorrhages in dogs [*54*] which received overdoses of intravenous amphotericin B indicated that a similar effect might be produced in man. Several patients receiving the antibiotic intravenously showed occult blood in the stool and in Case II there was one episode of gastrointestinal bleeding during treatment. This patient also had duodenitis and internal hemorrhoids, which may have been the sources of his blood loss. Nevertheless, the patient continued to show a gradual rise in hemoglobin while under treatment with the antibiotic.

Nine cases of systemic mycotic disease, other than coccidioidomycosis, were treated with amphotericin B by various routes by one of us (M. L. L.), in cooperation with other investigative groups. Reports of these cases will be published separately.

The tolerated doses of amphotericin B were as follows:

(1) *Intravenously*, 1.0 to 1.6 mg./kg., when administered on alternate days in 5 per cent glucose in distilled water over a six-hour infusion period.

(2) *Intra-articularly*, on alternate days, 25 mg. dissolved in sterile distilled water with streptokinase and streptodornase.

(3) *Intrathecally*, on alternate days, 0.7 mg. dissolved in sterile distilled water. A single intrathecal dose of 0.5 mg. of amphotericin B in 5 ml. distilled water produced a fungicidal level of 0.45 μg./ml. in the spinal fluid twenty-four hours later, disappearing, however, within forty-eight hours.

(4) *Intrapulmonary*, by aerosol inhalation, every six hours, 5 mg. dissolved in 1 ml. distilled water.

(5) *Intrathoracically*, in single doses of 3 mg.

dissolved in sterile distilled water. A single injection of 15 mg. of amphotericin B (an overdose) into the thoracic cavity of one patient, after pleuropneumonectomy, caused a severe febrile but non-fatal toxic reaction.

Maximum serum levels in patients receiving amphotericin B intravenously in a dosage of 1.2 mg./kg./day were usually no higher than 1.8 μg./ml. A small but significant difference was found between serum levels of the antibiotic and the minimal inhibitory concentration for C. immitis (0.5 μg./ml.), which would be quickly eliminated if the organism gained in resistance. The need for prolonged therapy of the systemic mycoses, moreover, would favor the development of resistance of the etiologic agent. Induced resistance to amphotericin B does occur, as demonstrated by Stout and Pagano [*66*], who noted that one of five strains of C. albicans became resistant; Littman and co-workers [*52*] also noted increase in resistance to amphotericin B by C. tropicalis, C. guilliermondii, C. krusei, C. stellatoidea and C. parakrusei. Some preliminary evidence that C. immitis does not become readily resistant to amphotericin B is found in the fact that a second isolate recovered in Case II, after thirty-two intravenous doses of antibiotic had been administered, was as sensitive to amphotericin B as the first isolate.

Although intravenous amphotericin B appears to have value in the specific treatment for coccidioidomycosis, other supportive forms of therapy, particularly surgical, should be employed in conjunction with it whenever indicated. These include pulmonary resection, surgical dèbridement and drainage of abscesses and sinuses, immobilization of affected extremities, control of secondary bacterial infection, and bedrest. Because of the relatively toxic nature of amphotericin B when given intravenously, its administration should be restricted solely to proved cases of systemic mycoses.

Prolonged therapy was deemed necessary because of the nature and chronicity of the disease. This was illustrated by the relapse encountered in Case II. Despite the favorable results to date, final evaluation of the clinical effectiveness of intravenous amphotericin B must await a more extended follow-up period. In view of the demonstrated effectiveness of intravenous amphotericin B for coccidioidomycosis, it is of great importance that the disease be recognized early, particularly in non-endemic regions, so that specific antibiotic therapy may be instituted early enough to lessen the severity and extent of residual lesions. Examples of this were encountered in Case I, a patient who had had chronic coccidioidal osteomyelitis of the right tibia for seventeen years before the true etiology was discovered; and in Case II, a patient with chronic coccidioidal infection of the left knee for five years before its true cause was ascertained.

The salient clinical, epidemiologic and laboratory features of coccidioidomycosis, as well as the characteristics of amphotericin B were reviewed.

SUMMARY

Amphotericin B, an actively fungistatic and fungicidal polyene antibiotic derived from a streptomycete, produced a beneficial clinical effect in four patients with coccidioidomycosis when administered intravenously. Toxic side effects of intravenous medication were minimized by slow administration in glucose solution on alternate days, and they subsided when the antibiotic was temporarily discontinued. Amphotericin B was administered by a number of routes, i.e., intravenously, intra-articularly, intrathecally, intra-pulmonary, intra-thoracically and orally. Prolonged intravenous therapy was deemed necessary because of the nature and chronicity of the disease.

The toxicity of amphotericin B when given intravenously to human subjects appeared primarily to affect the kidneys. Excessive or too frequent doses caused increasing azotemia which did not occur, however, when the antibiotic was given on alternate days or when smaller doses were employed. Toxic effects of intravenous medication were not observed on the heart, liver or bone marrow, and no neurologic or cutaneous symptoms were noted.

Blood serum levels of amphotericin B adequate for treatment of coccidioidomycosis, as assayed by a tube-dilution method with C. albicans, were produced by intravenous medication. A maximum blood serum level of 1.8 μg./ml. was obtained with intravenous amphotericin B given at doses of 1.2 mg./kg./day, which fell to 0.9 μg./ml. the following day without therapy. Eight human isolates of C. immitis were inhibited *in vitro* by 0.5 μg./ml. of amphotericin B. A fall in complement fixation titer for coccidioidomycosis occurred during and after therapy.

Amphotericin B given orally is absorbed too poorly from the gastrointestinal tract to produce assayable blood serum levels. It proved to be of no clinical value whatsoever in one patient with coccidioidomycosis.

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REFERENCES

1. Littman, M. L. and Zimmerman, L. E. Cryptococcosis, Torulosis or European Blastomycosis, pp. 1–205. New York, 1956. Grune & Stratton.
2. Smith, C. E., Whiting, E. G., Baker, E. E., Rosenberger, H. G., Beard, R. R. and Saito, M. T. The use of coccidioidin. *Am. Rev. Tuberc.*, 57: 330, 1948.
3. Mold and fungous diseases. A growing public health problem. *Consumer Reports*, 20: 45, 1955.
4. Forbus, W. D. and Bestebreurtje, A. M. Coccidioidomycosis: a study of 95 cases of the disseminated type with special reference to the pathogenesis of the disease. *Mil. Surgeon*, 99: 653, 1946.
5. Vital statistics—special reports: national summaries: mortality from each cause by color and sex. United States, 1955. National Office of Vital Statistics, U.S. Dept of Health, Education & Welfare, vol. 46, p. 43, 1956.
6. Maddy, K. T. Coccidioidomycosis of cattle in the southwestern United States. *J. Am. Vet. M. A.*, 124: 456, 1954.
7. Maddy, K. T. Coccidioidomycosis in a sheep. *J. Am. Vet. M. A.*, 124: 465, 1954.
8. Smith, C. E., Beard, R. R., Whiting, E. G. and Rosenberger, H. G. Varieties of coccidioidal infection in relation to the epidemiology and control of the diseases. *Am. J. Pub. Health*, 36: 1394, 1946.
9. Benninghoven, C. D. and Miller, E. R. Coccidioidal infection in bone. *Radiology*, 38: 663, 1942.
10. Willett, F. M. and Weiss, A. Coccidioidomycosis in southern California: report of a new endemic area with a review of 100 cases. *Ann. Int. Med.*, 23: 349, 1945.
11. Smith, C. E., Saito, M. T. and Simons, S. A. Pattern of 39,500 serologic tests in coccidioidomycosis. *J. A. M. A.*, 160: 546, 1956.
12. Kritzer, M. D., Biddle, M. and Kessel, J. F. An outbreak of primary pulmonary coccidioidomycosis in Los Angeles County, California. *Ann. Int. Med.*, 33: 960, 1950.
13. Schwarz, J. and Muth, J. Coccidioidomycosis: a review. *Am. J. M. Sc.*, 221: 89, 1951.
14. Baum, G. L. and Schwarz, J. Coccidioidomycosis: a review. *Am. J. M. Sc.*, 230: 82, 1955.
15. Smith, C. E. Epidemiology of acute coccidioidomycosis with erythema nodosum, ("San Joaquin" or "Valley Fever"). *Am. J. Pub. Health*, 30: 600, 1940.
16. Smith, C. E. An epidemiological study of acute coccidioidomycosis with erythema nodosum. *Proc. Sixth Pacific Sc. Congr., 1939*, 5: 797, 1942.
17. Smith, C. E. Parallelism of coccidioidal and tuberculous infections. *Radiology*, 38: 643, 1942.
18. Smith, C. E. Coccidioidomycosis. *M. Clin. North America*, 27: 790, 1943.
19. Smith, C. E. Recent progress in pulmonary mycotic infections. *California Med.*, 67: 179, 1947.
20. Smith, C. E. Diagnosis of pulmonary coccidioidal infections. *California Med.*, 75: 385, 1951.
21. Smith, C. E. and Baker, E. E. A summary of the present status of coccidioidal infection. *Weekly Bull. California Dept. Pub. Health*, 20: 113, 1941.
22. Smith, C. E., Beard, R. R., Rosenberger, H. G. and Whiting, E. G. Effect of season and dust control on coccidioidomycosis. *J. A. M. A.*, 132: 833, 1946.
23. Smith, C. E., Beard, R. R. and Saito, M. T. Pathogenesis of coccidioidomycosis with special reference to pulmonary cavitation. *Ann. Int. Med.*, 29: 623, 1948.
24. Smith, C. E., Saito, M. T., Beard, R. R., Kepp, R. McF., Clark, R. W. and Eddie, B. U. Serological tests in the diagnosis and prognosis of coccidioidomycosis. *Am. J. Hyg.*, 52: 1, 1950.
25. Smith, C. E., Saito, M. T. and Beard, R. R. Histoplasmin sensitivity and coccidioidal infection. I. Occurrence and cross-reactions. *Am. J. Pub. Health*, 39: 722, 1949.
26. Winn, W. A. Pulmonary mycoses—coccidioidomycosis and pulmonary cavitation. A study of ninety-two cases. *Arch. Int. Med.*, 87: 541, 1951.
27. O'Leary, D. J. and Curry, F. J. Coccidioidomycosis. A review and presentation of 100 consecutively hospitalized patients. *Am. Rev. Tuberc.*, 73: 501, 1956.
28. Moore, J. A., Murphy, J. D. and Ward, D. E., Jr. Lobectomy in coccidioidomycosis. Report of two cases. *J. Thoracic Surg.*, 18: 484, 1949.
29. Greer, S. J., Forsee, J. H. and Mahon, H. W. The surgical management of pulmonary coc-

cidioidomycosis in focalized lesions. *J. Thoracic Surg.*, 18: 591, 1949.

30. Cotton, B. H. and Birsner, J. W. Surgical treatment in pulmonary coccidioidomycosis. Preliminary report of thirty cases. *J. Thoracic Surg.*, 20: 429, 1950.
31. Forsee, J. H. and Perkins, R. B. Focalized pulmonary coccidioidomycosis. A surgical disease. *J. A. M. A.*, 155: 1223, 1954.
32. Cotton, B. H., Paulsen, G. A. and Birsner, J. W. Surgical considerations in pulmonary coccidioidomycosis. Report of 100 cases. *Am. J. Surg.*, 90: 101, 1955.
33. Carter, R. A. Coccidioidal granuloma: roentgen diagnosis. *Am. J. Roentgenol.*, 25: 715, 1931.
34. Carter, R. A. Infectious granulomas of bones and joints, with special reference to coccidioidal granuloma. *Radiology*, 23: 1, 1934.
35. Miller, D and Birsner, J. W. Coccidioidal granuloma of bone. *Am. J. Roentgenol.*, 62: 229, 1949.
36. Dykes, J., Segesman, J. K. and Birsner, J. W. Coccidioidomycosis of bone in children. *Am. J. Dis. Child.*, 85: 34, 1953.
37. Birsner, J. W. and Smart, S. Osseous coccidioidomycosis: A chronic form of dissemination. *Am. J. Roentgenol.*, 76: 1052, 1956.
38. Carter, R. A. The roentgen diagnosis of fungous infections of the lungs with special reference to coccidioidomycosis. *Radiology*, 38: 649, 1942.
39. Birsner, J. W. The roentgen aspects of five hundred cases of pulmonary coccidioidomycosis. *Am. J. Roentgenol.*, 72: 556, 1954.
40. Gifford, M. A. Coccidioidomycosis in Kern County, California. *Proc. Sixth Pacific Sc. Cong., 1939*, 5: 791, 1942.
41. Mead, C. I. Coccidioidomycosis in children. *J. A. M. A.*, 146: 85, 1951.
42. Emmett, J. Coccidioidin sensitivity among school children in Phoenix (skin test and x-ray survey). *Am. J. Pub. Health*, 42: 241, 1952.
43. Baker, E. E., Mrak, E. M. and Smith, C. E. The morphology, taxonomy, and distribution of *Coccidioides immitis*. Rixford and Gilchrist 1896. *Farlowia*, 1: 199, 1943.
44. Vogel, R. A. and Conant, N. F. *Coccidioides immitis* spherule antigen in a complement fixation test for experimental coccidioidomycosis. *Proc. Soc. Exper. Biol. & Med.*, 79: 544, 1952.
45. Friedman, L., Pappagianis, D., Berman, R. J. and Smith, C. E. Studies on *Coccidioides immitis:* morphology and sporulation capacity of forty-seven strains. *J. Lab. & Clin. Med.*, 42: 438, 1953.
46. Converse, J. L. Growth of spherules of *Coccidioides immitis* in a chemically defined liquid medium. *Proc. Soc. Exper. Biol. & Med.*, 90: 709, 1955.
47. Friedman, L., Smith, C. E., Roessler, W. G. and Berman, R. J. The virulence and infectivity of twenty-seven strains of *Coccidioides immitis*. *Am. J. Hyg.*, 64: 198, 1956.
48. Gold, W., Stout, H. A., Pagano, J. F. and Donovick, R. Amphotericins A and B, antifungal antibiotics produced by a streptomycete. I. In vitro studies, p. 579. Antibiotics Annual 1955–1956. New York, 1956. Medical Encyclopedia, Inc.
49. Vandeputte, J., Wachtel, J. L. and Stiller, E. T. Amphotericins A and B, antifungal antibiotics produced by a streptomycete. II. The isolation and properties of the crystalline amphotericins, p. 587. Antibiotics Annual 1955–1956. New York, 1956. Medical Encyclopedia, Inc.
50. Summary of information for clinical investigators. E. R. Squibb & Sons, Division of Olin Mathieson Chem. Corp., New Brunswick, N. J., October 28, 1955.
51. Dutcher, J. D., Young, M. B., Sherman, J. H., Hibbits, W. and Walters, D. R. Chemical studies on amphotericin B. I. Preparation of the hydrogenation product and isolation of mycosamine, an acetolysis product, p. 866. Antibiotics Annual 1956–1957. New York, 1957. Medical Encyclopedia, Inc.
52. Littman, M. L., Pisano, M. A. and Lancaster, R. M. Induced resistance of *Candida* species to nystatin and amphotericin B, p. 981. Antibiotics Annual 1957–1958. New York, 1958. Medical Encyclopedia, Inc.
53. Summary of information for clinical investigators. E. R. Squibb & Sons, Division of Olin Mathieson Chem. Corp., New Brunswick, N. J., March 6, 1957.
54. Fungizone for injection. Squibb amphotericin B desoxycholate complex for injection. E. R. Squibb & Sons, Division of Olin Mathieson Chem. Corp., New Brunswick, N. J., June 26, 1957.
55. Sternberg, T. H., Wright, E. T. and Oura, M. A new antifungal antibiotic, amphotericin B, p. 566. Antibiotics Annual 1955–1956, New York, 1956. Medical Encyclopedia, Inc.
56. Steinberg, B. A., Jambor, W. P. and Suydam, L. O. Amphotericins A and B: two new antifungal antibiotics possessing high activity against deep-seated and superficial mycoses, p. 574. Antibiotics Annual 1955–1956. New York, 1956. Medical Encyclopedia, Inc.
57. Louria, D. B., Feder, N. and Emmons, C. W. Amphotericin B in experimental histoplasmosis and cryptococcosis, p. 870. Antibiotics Annual 1956–1957. New York, 1957. Medical Encyclopedia, Inc.
58. Baum, G. L., Rubel, H. and Schwarz, J. Treatment of experimental histoplasmosis, p. 878. Antibiotics Annual 1956–1957. New York, 1957. Medical Encyclopedia, Inc.
59. Fiese, M. J. Treatment of disseminated coccidioidomycosis with amphotericin B: report of a case. *California Med.*, 86: 119, 1957.
60. Littman, M. L. Preliminary observations on the intravenous use of amphotericin B, an antifungal antibiotic, in the therapy of acute and chronic coccidioidal osteomyelitis. Conference on coccidioidomycosis. U. S. Dept. Health, Education & Welfare and Arizona State Health Department, Phoenix, Arizona, February 11–13, 1957.
61. Littman, M. L. Liver-spleen glucose blood agar for *Histoplasma capsulatum* and other fungi. *Am. J. Clin. Path.*, 25: 1148, 1955.
62. Littman, M. L. A culture medium for the primary isolation of fungi. *Science*, 106: 109, 1947.
63. Halde, C., Wright, E. T., Pollard, W. H., II, Newcomer, V. D. and Sternberg, T. H. The

effect of amphotericin B upon the yeast flora of the gastrointestinal tract of man, p. 123. Antibiotics Annual 1956–1957. New York, 1957. Medical Encyclopedia, Inc.

64. Larson, R. and Scherb, R. E. Coccidioidal pericarditis. *Circulation*, 7: 211, 1953.
65. Gore, I. and Saphir, O. Myocarditis. A classification of 1402 cases. *Am. Heart J.*, 34: 827, 1947.
66. Stout, H. A. and Pagano, J. F. Resistance studies with nystatin, p. 704. Antibiotics Annual 1955–1956. New York, 1956. Medical Encyclopedia, Inc.
67. Tarbet, J. E. and Breslau, A. M. Histochemical investigation of the spherule of *Coccidioides immitis* in relation to host reaction. *J. Infect. Dis.*, 92: 183, 1953.

Seminar on Liver Disease

Pathologic Aspects of Cirrhosis*

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DESPITE its well recognized morphological, physiological and clinical features, hepatic cirrhosis has been the center of controversy for many years. This is reflected in a series of group studies [*1–3*] devoted to the classification of cirrhosis. The problems involved have become more interesting, but also more complex, by virtue of the fact that cirrhosis has become the fourth most frequent cause of death in persons above forty years of age in this country [*4*]. There is increasing appreciation of the significance of geographic pathology, and a variety of methods for producing the disease as well as for study of the accompanying disturbances of blood flow have been developed. In the following pages an attempt is made to survey the pathophysiologic phenomena which characterize cirrhosis, to utilize them to arrive at a workable definition based upon clinicopathological correlation, and to develop a suitable basis for classification.

Anatomical Definition. Cirrhosis is defined as an alteration of the liver in which (grossly) the presence of nodules is associated with an increase of the connective tissue. Microscopic examination further discloses hepatocellular degeneration, necrosis and regeneration associated with chronic inflammation and fibrosis. This definition was accepted in 1931 by the International Society for Geographic Pathology [*1*]. To this definition is frequently added, distorted reconstruction of the lobular architecture [*5–7*].

The question has been raised whether cirrhosis is a uniform process [*8*] or develops over several different pathways [*9*]. There has been, moreover, considerable argument whether cirrhosis is a primary disturbance of the hepatic cells with reactive connective tissue formation [*5–7*] or a primary mesenchymal lesion, inflammatory in character, in which the epithelial alterations are secondary to the mesenchymal changes, particularly to the scarring [*10–12*]. The latter concept would identify cirrhosis with a chronic hepatitis, as reflected in Himsworth's suggestion [*10*] to discard the term cirrhosis and designate as fibrosis the sclerosed appearance of the liver, to which Mallory [*11*] also has given emphasis. This morphologic approach, however, fails to do justice to the functional problem of cirrhosis which is characterized by cardinal features not necessarily found in chronic hepatitis, namely, (1) reduced hepatic function, (2) portal hypertension, (3) ascites, and (4) tendency to progression. These aspects require consideration in any anatomical definition separating cirrhosis from other chronic hepatic diseases even of diffuse character. To evolve such a definition, pathophysiologic phenomena in cirrhosis deserve discussion.

PATHOPHYSIOLOGIC FEATURES

Hepatocellular Degeneration and Necrosis. The manifestations of injury of liver cells can be divided into those which result from the causes of cirrhosis and those which are induced by the cirrhotic process itself, independently of the original noxious agent.

The etiologic factors in cirrhosis may produce a variety of changes. (1) *Homogenous cytoplasmic coagulation and single cell necrosis*, such as are characteristic for viral hepatitis [*13*], occur in cirrhosis developing from hepatitis. The response to the single cell necrosis, namely accumulation of mononuclear cells and circumscribed collapse of the framework, provides additional but even less specific criteria of viral hepatitis. (Fig. 1.) Surprisingly enough, such morphological criteria are found only exceptionally in cirrhosis. They may be seen only in postnecrotic cirrhosis adjacent to massive collapse. In other types of cirrhosis, especially those

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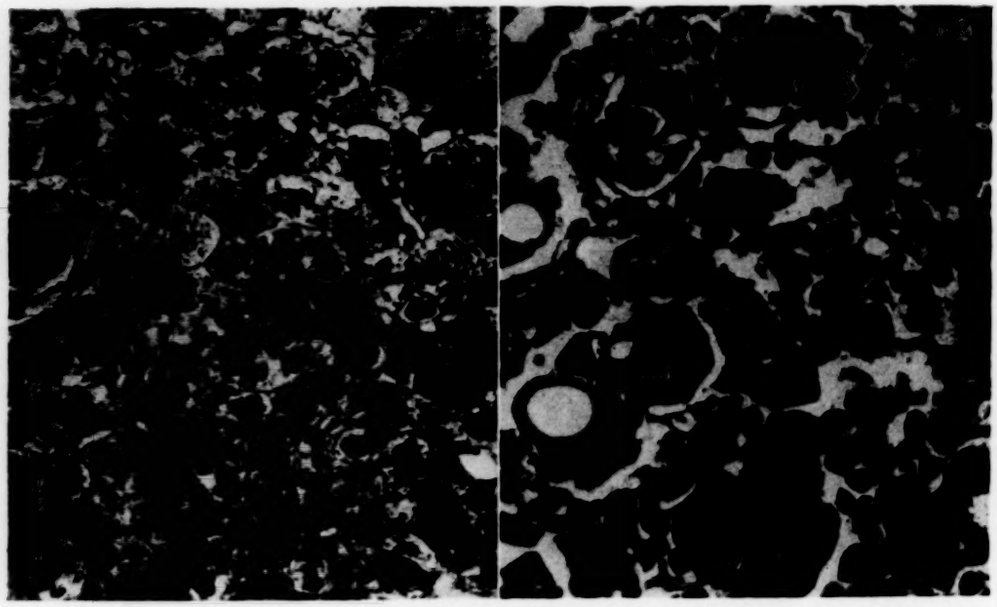


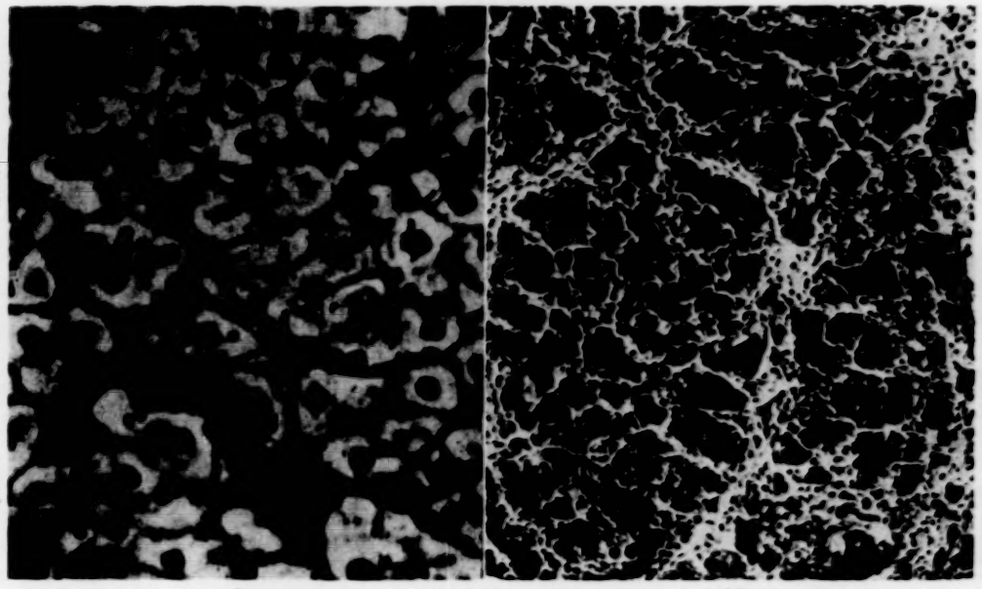
1A 1B

FIG. 1. A, acute viral hepatitis. Single cell necrosis shown by cluster of inflammatory cells (arrow). Diffuse degeneration of liver cells with focally accentuated regeneration (binucleated cells). Hematoxylin and eosin, × 430. B, florid cirrhosis in alcoholic subject. Numerous dark-staining ramified cytoplasmic coagulates (Mallory bodies) in hepatic cell cytoplasm (arrow). Mallory's aniline blue stain, × 430.

associated with malnutrition, these changes have never been observed by the authors. Viral hepatitis is either not an aggravating factor in cirrhosis or, as seems more likely, its morphologic manifestations are modified to a high degree. (2) *Focal granular clumping of the cytoplasm* is usually observed in swollen cells with rarified cytoplasm. The clumps aggregate to form a ramified perinuclear acidophilic body which has been designated by Mallory [*11*] as "alcoholic hyaline" and considered a specific effect of alcoholic intake. (Fig. 1B.) Similar lesions have been found in cirrhosis associated with malnutrition in the absence of alcoholism. Focal coagulation necroses in experimental poisoning, such as that produced by bromobenzene [*14*], are histochemically different and thus the assumption that the lesion is specific is justified [*15*]. This form of degeneration is associated with focal aggregation of segmented leukocytes. (3) *Centrolobular necrosis* occurs in chemical poisoning, other types of toxic hepatic injuries and passive congestion.

Lesions of the liver cells induced by the cirrhosis itself and not by its original cause result either from disturbance of flow of blood or of bile: (1) *Ischemic or hypoxic degeneration and necrosis* develop in the center of the nodule. They greatly resemble toxic or congestive necrosis, indicating that the underlying cause probably is oxygen deficiency interfering with enzymatic processes [*17,18*]. (2) *Topical effects of bile upon hepatic cells* resulting in a reticular arrangement and bile pigmentation of hepatic cytoplasm (feathery degeneration) is found in all types of cholestasis associated with cirrhosis. In a peculiar sub-variety resulting from prolonged cholestasis, the cells on the periphery of the lobule disintegrate. In primary or secondary biliary cirrhosis this process interrupts the communication of liver cells with bile ductules. Subsequent fibrosis makes this interruption permanent. This mechanical biliary obstruction may persist even if a concomitant obstruction of the extrahepatic biliary tract has been relieved surgically.

Massive necrosis of all cells in one lobule or in contiguous parts of several lobules may be the result either of the original cause of the cirrhosis or of the cirrhotic process itself. In contrast to single cell necrosis, it is not readily repaired by regeneration but leads to massive or submassive collapse. Necrosis of groups of cells may be followed either by regeneration or by focal collapse of stroma. (Fig. 2A.) The collapsed areas may produce mechanical stress on the surrounding parenchyma with resulting break fissures



2A 2B

FIG. 2. A, focal intraparenchymal collapse of stroma shown as area of condensed reticulum. Gomori's reticulum impregnation, × 430. B, "giant cell hepatitis of infancy" showing numerous large multinucleated liver cells and early cirrhosis. Hematoxylin and eosin, × 100.

which subdivide the adjacent uninvolved parenchyma [9]. Massive collapse may affect previously normal parenchyma. This *primary* collapse is characterized by normally arranged but approximated pre-existing vessels which outline denuded ghost lobules. Previously cirrhotic nodular parenchyma may also undergo massive necrosis. In the resulting *secondary* collapse, the arrangement of the vessels is entirely irregular. Experiments with ethionine-induced cirrhosis have shown that massive collapse does not always result from massive necrosis associated with obvious clinical manifestations but may also develop from repeated single cell necrosis not necessarily associated with functional alterations [22]. At laparotomy or autopsy rather extensive collapse frequently is noted in patients with surprisingly little functional and clinical abnormality.

Hepatocellular degeneration in cirrhosis is mirrored by malaise, anorexia, hemorrhagic diathesis and particularly by endocrine manifestations. Hepatic tests reflect these hepatocellular changes fairly well [19,20], especially the cholesterol ester/cholesterol ratio, cephalin flocculation test, thymol turbidity test, serum gamma globulin elevation and particularly the serum albumin level [20,21], although some investigators emphasize, in contrast, the lack of correlation [25]. Some correlation with the serum mucoprotein level also has been noted [27], and recently elevation of the serum transaminase has been considered to be an index of hepatic dysfunction in cirrhosis [28].

Fatty Metamorphosis. Fatty metamorphosis may precede cirrhosis, as in alcoholism or chronic malnutrition, and then may represent, at least in the opinion of some, the cause of cirrhosis. In other instances, however, fatty metamorphosis complicates any type of cirrhosis, being secondarily induced by nutritional disturbances in the course of the disease. In animals with choline deficiency, fat accumulates in the center of the lobule, while in protein deficiency fat accumulates predominantly at the lobular periphery [26,29–32]. In experimental animals, particularly rats and mice, fatty metamorphosis as such induces cirrhosis. Some believe that the same holds true for man [10,33] whereas others [34,35], especially in England [36], deny a direct causal relation between fatty metamorphosis and cirrhosis. Differences in the vascular supply between human and animal livers [37] can explain the tendency of the rat liver to become cirrhotic as the result of fatty metamorphosis alone.

Accumulation of an abnormal amount of fat in the liver, when the result of nutritional imbalance or endocrine disturbances (e.g., in diabetes, obesity), is not necessarily reflected in

alteration of the results of hepatic tests [*38*]. Even in advanced fatty metamorphosis, in which the microscopic picture resembles adipose tissue and suggests a conspicuous reduction of hepatic cytoplasm, liver function appears surprisingly well preserved except for increased bromsulphalein retention. The major alteration established biochemically is a reduction of DPN in the cells [*39*]. However, a tendency to fibrosis does result from excessive accumulation of fat.

Inflammatory Reaction and Splenomegaly. Inflammation in cirrhosis may be a reaction to the original cause of cirrhosis, as in viral hepatitis, parasitic infections and granulomatous diseases, or it may be secondary to hepatic necrosis. For instance, in viral hepatitis, it is not established to what degree the parenchymal inflammation is a reaction to the virus or to liver cell breakdown. In all forms of cirrhosis, inflammatory cells accumulate, probably as a response to liver cell injury. The basophilic cytoplasm of some of these mononuclear inflammatory cells and of the proliferated Kupffer cells is rich in nucleoprotein as judged by the pyroninophilia specifically removed by ribonuclease. Since cytoplasmic nucleoproteins are associated with protein formation, it has been assumed by some that the mesenchymal cells of the liver form the excess serum gamma globulin frequently observed in patients with cirrhosis. However, in coarse nodular cirrhosis experimentally induced by ethionine the elevation of serum gamma globulin is related to the basophilic cells of spleen and lymph nodes, suggesting that the latter are at least in part responsible for the hypergammaglobulinemia. In the white pulp of the spleen and in the medullary cords of the lymph nodes these pyroninophilic cells, morphologically not identical with plasma cells but presumably related to them, are greatly increased in number. After splenectomy the hypergammaglobulinemia of the ethionine intoxicated rats is not reduced but the lymph nodes are even more strongly pyroninophilic, suggesting their compensatory role under such conditions [*40*]. Spleen and lymph nodes from patients with cirrhosis associated with hypergammaglobulinemia likewise show increased numbers of these basophilic (pyroninophilic) cells [*41,42*]. Additional experiments are necessary to elucidate whether or not this plasmacytoid response is due to sensitization by hepatic cell breakdown products. Some types of necrosis, especially in postnecrotic cirrhosis, might represent a reaction to antibodies against liver tissue. The possibility of such a reaction has recently been raised by the demonstration of an L.E. cell phenomenon [*43*] and of antibodies against liver in patients with chronic hepatitis [*44*].

The splenomegaly in cirrhosis is usually considered the result of the hydrostatic pressure produced by portal hypertension [*45*]. However, part of it results from reactive hyperplasia of the pulp independent of portal stasis. This is exemplified in ethionine cirrhosis in which the spleen is enlarged to ten times its original size. Most of this must be attributed to the pulp cell hyperplasia which is related to the activity and duration of the hepatocellular process. With subsidence of the acute intoxication and damage to liver cells there is a fall in gamma globulin levels and a decrease in splenic enlargement to about three times its original weight; the persistent splenomegaly correlates with the degree of stabilized cirrhosis [*40*].

Regeneration. Replacement of liver cells by regeneration, a process which normally takes place but at a rate so far unknown, becomes accentuated in the vicinity of focal or submassive necrosis, regenerating cells growing into and expanding the framework. The interruption of the liver cell plates seemingly provides a growth stimulus. Hepatic regeneration may also be conspicuous in intact parenchyma, especially in the periphery of the lobules and distant from the site of liver cell loss, apparently under a humoral influence which removes a growth inhibiting factor [*46,47*]. It can also be elicited by preparations containing nucleic acid and by liver tissue implantation [*48*]. Morphologically, regeneration may be reflected in cytologic changes such as increased cytoplasmic basophilia and nuclear and nucleolar enlargement and multiplication (polyploidy) [*49*], as well as by architectural alterations such as formation of liver cell plates more than one cell thick. Nuclear division may outdistance cytoplasmic growth and division, resulting in multinuclear giant cells (Fig. 2B), especially in the infantile liver. Giant cell cirrhosis and hepatitis is therefore not an entity [*50*]; neither is it a reflection of a specific disturbance in bile capillary formation but rather an expression of the increased regenerative ability of the infantile liver occurring in congenital syphilis, viral hepatitis, extrahepatic biliary obstruction and blood group incompatibility.

Both cytologic and architectural regeneration in intact parenchyma takes place mainly in the periportal zone. This is seen in animals on carcinogenic diets before the appearance of hepatic cancer. Accentuated regeneration in a circumscribed area initiates the formation of nodules. Groups of liver cells initially show cytological characteristics of regeneration without interruption of the lobular architecture. When the liver cell plates become more than one cell thick, they become interrupted. Nodular hyperplasia exerts pressure upon the surrounding parenchyma and stroma. Eventually this stroma, after loss of most of its liver cells, becomes compressed by the "active" nodule into a "passive" connective tissue septum. (Fig. 3.) Active nodule formation may start from cells isolated in the connective tissue after necrosis or separation by a septum. The periphery of an active nodule frequently shows marked cytologic and architectural regeneration [*9*]. In the nodules the liver cells lose their original radial alignment; instead they converge towards the center of the nodule where sinusoids become transformed into an efferent vein. As the nodule grows, portal vein branches and bile ducts grow into it to produce a secondary "lobulization," as observed in rats with chronic ethionine intoxication [*51*]. Before this "lobulization," the regenerative nodule may show a certain metabolic independence by exhibiting differences in fat or glycogen content and in the type and number of inflammatory cells. These differences tend to disappear with increasing lobulization. Lobulization may eventually lead to secondary formation of normal central veins and portal tracts which, however, are smaller [*52*]. It is therefore possible that, especially in postnecrotic cirrhosis, seemingly normal parenchyma is actually newly formed. The difference in color and appearance of isolated nodules may be the result of local necrosis from disturbances of blood flow or focal bile stasis but usually reflects metabolic autonomy which makes such nodules susceptible to malignant change. If changes of this kind are observed a thorough search for fully developed carcinoma should be made in other parts of the liver. Despite the widely held belief of the unicentric origin of cancer [*53*], a multicentric origin is the rule in experimental hepatic carcinoma and frequently in cirrhosis in man [*54*]. Its relation to regeneration is reflected in a high incidence in postnecrotic cirrhosis [*55,56*] and occurrence in juvenile hepatitis [*57*].

FIG. 3. Experimental ethionine poisoning of rat. Several "active" nodules are compressing "passive" septums. Gomori's reticulum impregnation, × 100.

Ductular Cell Reaction. The bile canaliculi are lined by an enforcement of the liver cell membrane which under the electron microscope appears as a series of fine protoplasmic projections [*58*]. (Fig. 4A.) The canaliculi are connected with the septal bile ducts in the portal tracts by tubules which occur predominantly in the periportal zone but also intralobularly, and for which such names as cholangioles, canal of Hering, intermediate pieces, etc. have been coined but which are best designated bile ductules. The ductular cells vary in size and are cuboidal as a rule. They have fewer mitochondria than the liver cells when studied under the electron microscope but have the same luminal cytoplasmic projections. While normal liver cells have microvilli at their base extending into a space freely connecting with the bloodstream and not exhibiting a basement membrane, the ductular cells have no basal microvilli but a basement membrane. (Fig. 4B.) Embryonically, the ductular cells seem to derive from the liver cells rather than vice versa [*59*]. In many abnormal conditions, particularly in cirrhosis, excessive proliferation of ductular cells

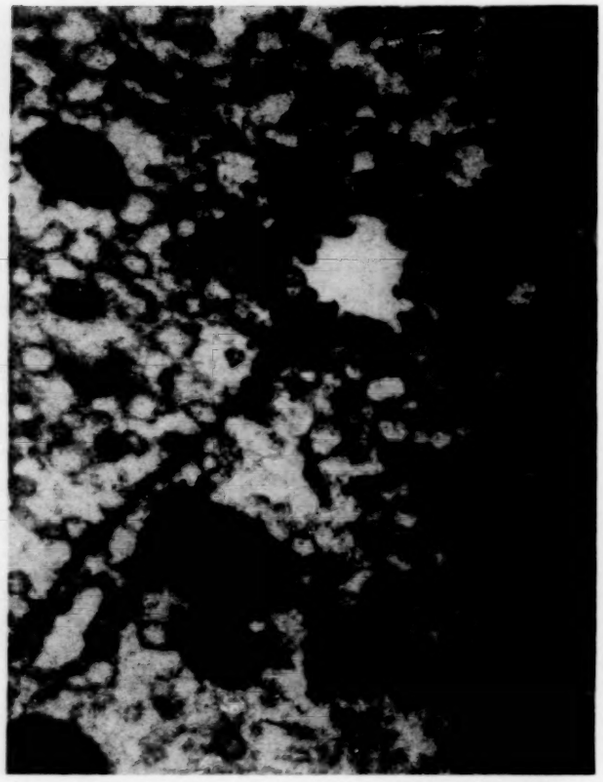


FIG. 4A. Electronmicrograph of frog liver. Cytoplasmic villi of hepatic cells protrude into lumen of bile canaliculus (arrow). × 17000 (Provided by Dr. W. Mautner).

FIG. 4B. Electronmicrograph of rat liver. Several ductular cells with rather small mitochondria. Of note are cytoplasmic luminal villi. × 6000 (Provided by Dr. W. Mautner).

is noted. These cells may be arranged in readily recognizable ductules or singly or in clumps. Their nature became more clarified as the result of a study of ethionine intoxicated rats. In this condition, the liver cells are surrounded by clusters of interstitial cells. Some of these are unquestionably inflammatory while others have been referred to as oval cells [*60*] (Fig. 5) and considered to be hyperplastic bile duct epithelium. Similar cell groups have been found in other conditions such as experimental biliary obstruction and after exposure to a variety of hepatic carcinogens [*60–63*]. The non-inflammatory interstitial cells differ from Kupffer's cells by their lack of phagocytosis when injected with India ink. In chronic intoxications ductular cells may become more elongated and are arranged in single file, resembling fibroblasts. Their ductular nature can be demonstrated not only by a positive histochemical reaction for alkaline phosphatase but particularly by injection of the biliary tree with India ink which discloses finely branching channels within the interstitial clusters of apparent mesenchymal cells [*64*]. In very thin sections, a distinct cuticular luminal membrane can be seen. Similar ductular cells are found in the human liver, likewise frequently associated with inflammatory cells, particularly in the lobular parenchyma where they may suggest reaction to focal necroses in both non-specific reactive hepatitis or in viral hepatitis. These cells also contribute to the cellularity of the portal tracts in various inflammatory conditions and are frequent in the trabeculae of cirrhosis. Doubtless, many of these excess ductular cells derive from biliary epithelium. However, the possibility cannot be excluded that intralobular ductular cells are derived from liver cells, as in the course of embryonic development, especially when they are arranged in plates rather than cords [*65*]. Whatever its origin, the ductular cell reaction is a response of the liver to injury of various kinds. Together with the inflammatory reaction, the ductular cell reaction is suppressed by cortisone in ethionine intoxication, although the damage to liver cells is not influenced by cortisone [*66*]. A basement membrane can be demonstrated giving periodic acid-Schiff (PAS) reaction

around both organized and disorganized ductular cells; neighboring Kupffer's cells contain similarly stained granules, possibly a precursor material [*67*]. This basement membrane is enforced by reticulum fibers which later in their development give collagen staining reactions. These reticulum fibers surround the ductular cells, sometimes in multiple layers. The formation of ductular cells thus represents a fibroblastic stimulus. The role of the ductular cell in stimulating or contributing to fiber formation needs further study.

HISTOGENESIS OF FIBROSIS

The hepatic connective tissue framework consists of (in addition to elastic fibers and membranes mostly in vessels) (1) reticulum fibers appearing black with silver impregnation and under favorable circumstances as purple with the PAS reaction, and (2) collagenous fibers and membranes demonstrable by a variety of staining procedures, e.g., the van Gieson reaction or Mallory's aniline blue procedure and appearing brown following silver impregnation. These fibers normally are rather thick and appear in sections as comma-shaped structures in the portal tract whereas the few membranes present appear as thin wavy lines [*68*] extending into the parenchyma. Collagen and reticulum apparently are closely related. Despite morphologic differences [*69*], both fibrils, when studied with the electron microscope, show a characteristic periodicity of 640 Å [*70,71*]. Both contain considerable amounts of hydroxyproline, an amino acid present in other proteins only in very small amounts. Liver in man, which is much richer in reticulum than the rat liver, contains three times as much hydroxyproline [*72*]. Renal reticulum not only contains much less hydroxyproline than collagen but also is admixed with a lipoglycoprotein [*73*], the carbohydrate moiety being reflected in the positive PAS reaction [*74*]. There is evidence for the existence of different reticulins [*75*]. Preliminary studies have indicated that newly formed reticulum in experimental cirrhosis is richer in hydroxyproline [*72*]. In human and experimental cirrhosis the content of hepatic collagen and reticulum, chemically estimated, is considerably elevated [*76*]. Moreover, at least in experimental cirrhosis, an interfibrillary PAS-positive ground substance has been demonstrated [*77*] which gives staining reactions for acid mucopolysaccharides. Collagen and reticulum are far less sensitive to anoxia, viral or bacterial infection or chemical poisons than the liver cells. The stroma consequently persists in most forms of hepatic injury and disruption of the reticulum framework is rare, occurring principally in infarcts.

FIG. 5. Experimental ethionine poisoning of rat. Interstitial cell clusters composed of ductular cells intermixed with few leukocytes surround large liver cells. Hematoxylin and eosin, × 585.

Increased fibrous tissue in the human liver may be the result either of (1) approximation (collapse) of preformed stroma after disappearance of the parenchymal cells, or (2) new formation, or (3) both.

Collapse. After massive necrosis or after continued focal necrosis, the reticulum framework becomes approximated and the larger vessels consequently lie much closer to each other. When this process develops in the normal liver (primary collapse), the basic arrangement of the portal tracts and central fields is preserved and the original borders of the portal tracts are easily discernible. When necrosis ensues in cirrhotic parenchyma (secondary collapse), the arrangement of the vessels is irregular and the border of the original portal tracts cannot be identified [*9*]. After necrosis of exten-

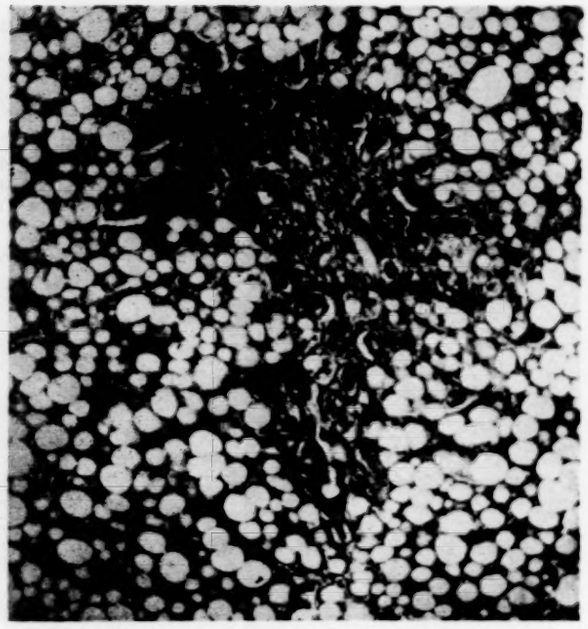


FIG. 6A. Fatty liver of alcoholic subject. Star-shape of enlarged portal tract produced by radiating collagenous membranes and septums. Hematoxylin and eosin, × 100.

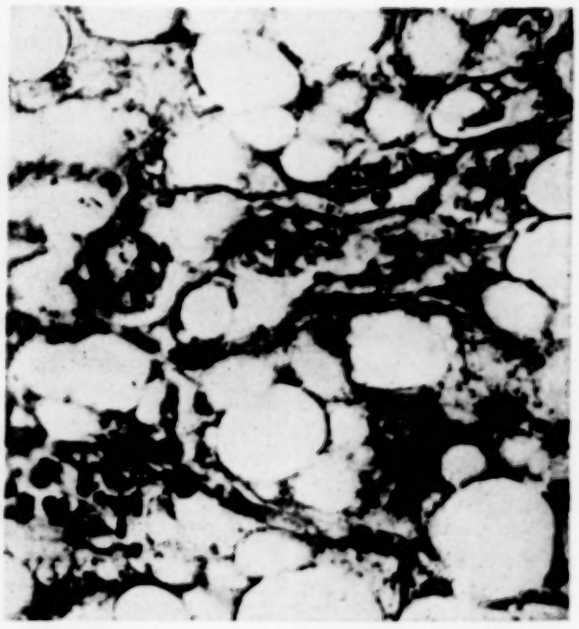


FIG. 6B. Fatty liver of alcoholic subject. Collagenous membranes with duplication around fatty liver cells and fatty cysts. Mallory's aniline blue stain, × 430.

sive parts of several lobules, submassive collapse takes place; occasionally, particularly in viral hepatitis, only small portions of the lobule may appear collapsed.

New Formation of Fibers. Fibers or membranes may form in the liver with or without conspicuous appearance of fibroblasts. (*1*) *Formation of fibers in the presence of fibroblasts* takes place from Glisson's capsule [*78*] and also in granulation tissue associated with nonspecific or specific inflammatory processes. This includes the reaction around some forms of extracellular fat or escaped bile. It is frequently found, therefore, in chronic biliary obstruction and especially in infants with biliary atresia. This type of fibrosis has been experimentally produced by implantation of foreign bodies [*79*] or intrahepatic injection of carrageenin [*80*]. Under all these circumstances the reticulum and collagen content is increased and transition from the former to the latter may be assumed. In the carrageenin experiments, the fibroblasts are rich in PAS-positive material, possibly the precursor of the reticulin [*81*]. This type of fibrosis is apparently the result of a primary stimulation of portal mesenchyma without participation of hepatic epithelium. Hepatic fibrosis without extensive reticulum formation occurs in scirrhous carcinoma and Gaucher's disease where it is associated with fibroblasts and represents a primary type of scarring. It also occurs in cirrhosis following granulomatosis, cholangitis or suppurative hepatitis. (*2*) *New formation of fibers without conspicuous accumulation of fibroblasts.* Fibroblasts are not frequent in the common form of cirrhosis associated with fatty infiltration in alcoholic persons but have been demonstrated in fatty cirrhosis experimentally produced by choline deficiency [*82*].* An increase of reticulum fibers, subsequently undergoing "collagenization," occurs in association with new formation of ductular cells. This is especially conspicuous on the border of portal tracts where liver cell plates connect with ductules possibly as an expression of the transformation of liver cells into ductules. This results in the formation of reticulum fibers and subsequent collagenous membranes which radiate from the portal tracts into the parenchyma, a feature characteristically seen in active stages of various types of cirrhosis, for example, those associated with malnutrition and fatty metamorphosis, and in hepatitis and acute stages of hemochromatosis or in prolonged cholestasis. These fibers aggregate to form septums which either enlarge the portal tract diffusely or aggregate only on a part of the periphery of the portal tracts or central canals to produce a stellate appearance. (Fig. 6A.) Frequently, duplicated basement

* The role of the Kupffer cells requires clarification.

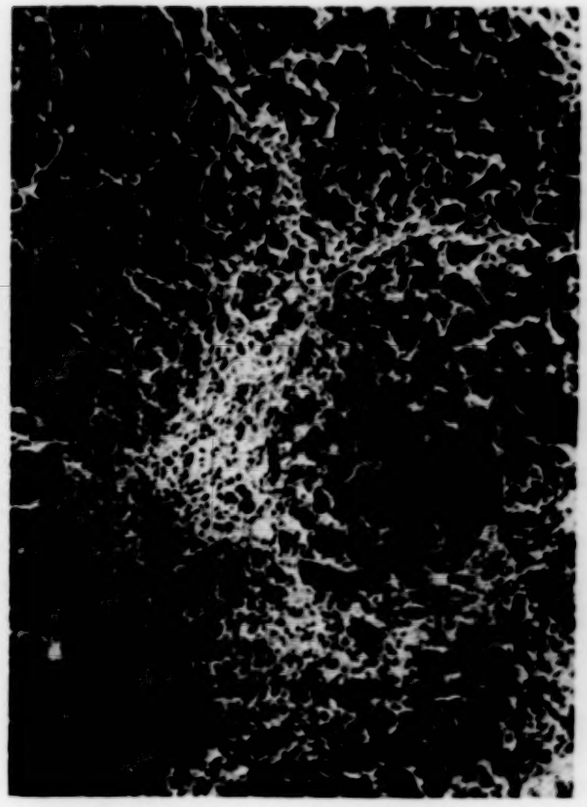


FIG. 7. Centrolobular collapse and fibrosis with beginning regeneration of parenchyma. Hematoxylin and eosin, × 75.

FIG. 8. Periportal fibrosis and inflammation with partial blurring of parenchymal borders. Hematoxylin and eosin, × 80.

membranes (Fig. 6B) can be recognized, by reticulum impregnation and subsequently by collagen stains, around liver cells containing much fat and especially those which border fatty cysts derived by coalescence of fat droplets from several cells [*83*]. Membranes of the same nature border liver cells exhibiting degeneration, such as coagulation necrosis or Mallory's "hyaline." A similar increase in reticulum develops also in areas of passive congestion and sometimes near hepatic necrosis. If the ductular cell reaction is considered an expression of dedifferentiation of liver cells in response to injury, and if the lack of (electron microscopically visible) cellular projections of the ductular cells toward the bloodstream also is considered to be related to this dedifferentiation, then in certain liver cell alterations a similar dedifferentiation also may be associated with the formation of a basement membrane proceeding to fibrosis.

Collapse Combined with New Formation of Connective Tissue. In prolonged primary and secondary massive, submassive or focal collapse, the arrangement of the reticulum fibers is altered and "collagenization" of some fibers takes place [*9*], resulting at first in thin and later in thick membranes traversing the collapsed area and communicating with portal tracts and central fields, the outlines of which become blurred. Fibroblasts are inconspicuous. After several months the hydroxyproline content seems to increase [*72*]. Some of the collagenous fibers are arranged around dilated sinusoids which are transformed into veins anastomosing with portal and hepatic canals. Fibrosis resulting from such collapse is irregularly shaped.

Long, thin, straight areas of fibrosis consisting of collagenous membranes intermingled with reticulin fibers are not related to the lobular architecture and appear on three-dimensional reconstruction as septum [*9*]. These develop apparently in break fissues which form either in the vicinity of a rapidly developing massive collapse or separate territories of hepatic parenchyma of different tissue turgor, for instance, as the result of conspicuous fatty metamorphosis and regeneration separated from areas of either necrosis or atrophy [*9*]. These break fissures

FIG. 9. Periductular fibrosis near portal tract (left hand margin) extending into parenchyma. Mallory's aniline blue stain, × 100.

FIG. 10. Septums subdividing parenchyma in cirrhotic liver with fatty change. Mallory's aniline blue stain, × 100.

evolve from mechanical stress. In such fissures the individual liver cells have disappeared and sinusoids are transformed into veins in the wall of which collagen membranes develop, possibly in relation to the blood pressure (law of Thomas). Moreover, around focal necroses or granulomas, neoformation of fibers, induced either by liver cell degeneration or fibroblastic proliferation, may occur in association with disappearance of liver cells. It has also been claimed [*84*] that new formation of elastic fibers is a characteristic feature of cirrhosis.

MORPHOLOGY OF FIBROSIS

Excess fibrous tissue may present itself in the following configurations: (1) *Focal intraparenchymal fibrosis* (Fig. 2A) follows extensive focal necrosis, especially when associated with granulomas. (2) *Centrolobular fibrosis* (Fig. 7) occurs particularly after passive congestion and presumably following intoxication by certain drugs. (3) *Portal fibrosis* develops after portal inflammations such as non-specific pericholangitis, extrahepatic biliary obstruction and various granulomatous diseases including schistosomiasis. (4) *Periportal fibrosis* (Fig. 8) extends into the adjacent parenchyma as (a) diffuse widening of the portal tract, as seen in chronic biliary stasis, cholangitis, and also in hemochromatosis, or (b) stellate enlargement produced by extension of short septums into various directions from the portal tract, seen in beginning nutritional cirrhosis with fatty change, kwashiorkor and chronic viral hepatitis. (5) *Periductular fibrosis* (Fig. 9) develops around proliferating ductules in the periportal zone and within the lobule when fibers accumulate following inflammation, probably the result of regurgitation of bile in intra- and extrahepatic cholestasis [*85*] and possibly following primary pericholangiolitis [*86*]. (6) *Septal fibrosis* (Fig. 10) manifests itself in septums traversing the lobular parenchyma with or without relation to the lobular architecture. They appear as straight or irregularly shaped bands of varying width which in three-dimensional reconstruction [*9*] are septums or trabeculae. They may either surround the lobules in the form of perilobular fibrosis (Fig. 11) [*23*] or extend into it as advanced stellate fibrosis or as result of stress fissures. These septums, irrespec-

FIG. 11. Perilobular fibrosis. Mallory's aniline blue stain, × 110.

tive of origin, may connect with each other and also with foci of intralobular fibrosis [87]. Their arrangement has also been associated with the shape of an acinar unit which overlaps that of the lobule [88]. The end result may be a septal connection between the portal and central canals. Since these canals cross or interdigitate in space, the "active" septums connecting them may obliterate part of the lobule to form irregularly shaped "passive" nodules. (Fig. 12.)

(7) *Postcollapse fibrosis* (Fig. 13) results from submassive and massive collapse and is characterized by broad connective tissue bands which are apparent on gross examination.

CHANGES OF HEPATIC VASCULATURE

Injection technics of isolated organs have demonstrated characteristic alterations of the vascular tree of the liver in cirrhosis, some of which may also be shown by splenoportal radiography during life [89]. After injection of portal and hepatic vessels in cirrhosis with plastics, followed by digestion of the tissue, a cast of the vascular tree is obtained. Under these circumstances the portal vein branches of cirrhotic subjects, especially when associated with ascites, show deformities and irregular arrangement; the arteries appear normal [90]. The hepatic vein tributaries are not as well protected by surrounding connective tissue and by hepatic arteries and bile ducts as the portal vein branches, consequently they are compressed where they cross between regenerative nodules [91,92] and are distorted by fibrosis [93]. This postsinusoidal compression interferes with the drainage of blood from the liver and is thus one of the main causes of portal hypertension in cirrhosis [91,92]. Since smaller nodules compress veins more effectively than larger ones, they are associated with a greater tendency to portal hypertension [94]. The contribution of the hepatic arteries to the nodules is greater than that to the normal parenchyma [93]. Arterial branches likewise contribute more to the vasculature of primary or secondary hepatic carcinoma [95].

FIG. 12. Schematic drawing of septums extending between portal and central canals which cross in space. (From POPPER, H. In: Liver Injury, Trans. Eleventh Conf., 1952, New York. Josiah Macy, Jr. Foundation.)

If the portal and hepatic vessels are injected with gelatin of different colors, tissue sections of the liver show the septums injected from all three vessels [92] while in animals with carbon tetrachloride cirrhosis the septums are injected chiefly from the hepatic vein [96]. In many places parasinusoidal communications between branches of the portal and hepatic veins are noted which shunt blood from the former into

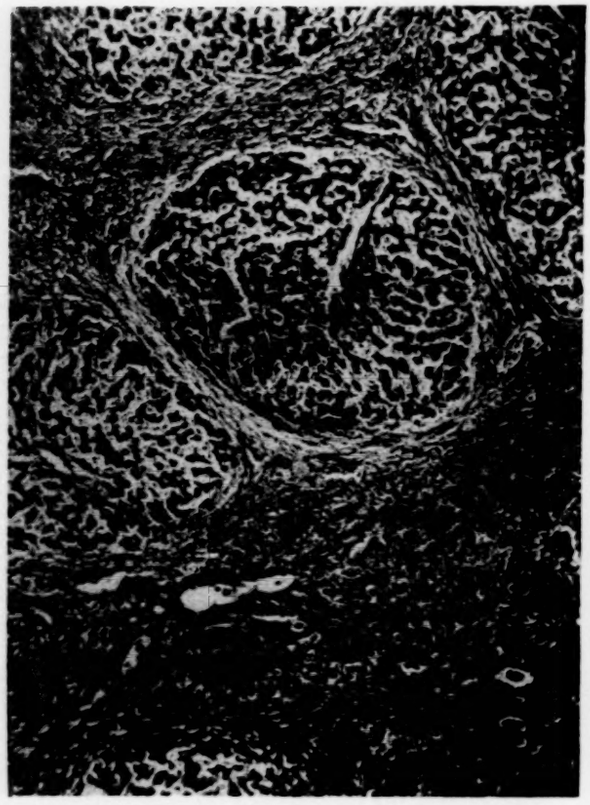


FIG. 13. Postnecrotic cirrhosis with broad bands resulting from collapse and nodules with intact lobular architecture. Hematoxylin and eosin, × 43.

FIG. 14. Septum containing veins extending from portal tract (left hand margin) separating regenerative nodules characterized by plates two cell thick; one of them is separated by a break fissure (arrow) from non-regenerative paranchyma. Hematoxylin and eosin, × 100.

the latter, bypassing the parenchyma [*92,93*]. Together with extrahepatic portosystemic (and portopulmonary [*97*]) anastomoses, they serve to divert blood from the hepatic parenchyma. In experimental acute hepatic injury, a similar partial portohepatic venous shunt can be demonstrated in that injection material passes rapidly from the portal to the hepatic vein branches through a few sinusoids, avoiding the great majority [*64*]. The functional efficiency of the portohepatic anastomoses is disclosed by the tendency to bacteremia with enterobacteria in severe liver disease [*98*], in the development of urate calculi in the urinary tract in experimental carbon tetrachloride cirrhosis [*99*], and in the rarity of metastatic carcinoma in livers from cirrhotic subjects [*100*]. Cancer in livers from cirrhotic subjects is much more likely to be primary in origin.

Presinusoidal anastomoses between portal vein and hepatic arterial branches also are present in cirrhotic livers [*92,93*]. Through these anastomoses arterial pressure is brought to bear upon the portal venous system to contribute to the portal hypertension. This component of portal hypertension is revealed in the measurements of the pressure in the proximal stump of the portal vein after its separation during shunt operation in cirrhotic subjects.

Reduction of blood flow through the hepatic parenchyma in cirrhosis has been demonstrated by bromsulphalein extraction [*101,102*] and the rate of disappearance of radioactive colloidal gold [*103,104*]. Reduction of the estimated hepatic blood flow in cirrhosis is accompanied by an increase in the arteriovenous oxygen difference, suggesting increased hepatic utilization of oxygen. This does not correlate well with the demonstrable portohepatic anastomoses, a discrepancy possibly explained by a retarded blood flow in the portal system. In fatty livers without cirrhosis, the blood flow is not necessarily reduced although the arteriovenous oxygen difference is increased [*105*], possibly also because of retardation of presinusoidal blood flow.

DEFINITION OF CIRRHOSIS BASED ON STRUCTURAL-FUNCTIONAL CORRELATION

A valid connotation applicable to both functional and structural characteristics of cirrhosis has to take into account the clinical features reflected in (1) portal hypertension, (2) reduced hepatic function, (3) tendency to progression of the disease, and (4) ascites. This can be accomplished by considering the anatomic characteristics of cirrhosis, the alterations associated with reconstruction of the lobular architecture as reflected in two specific features: (1) the regenerative nodule and (2) the connective tissue septum linking portal tracts with central canals [*9,106*]. (Fig. 14.)

The regenerative nodules, by producing postsinusoidal obstruction, are responsible for most of the portal hypertension in cirrhosis. The vascular anastomoses in the septums, specifically the presinusoidal communications between portal vein and hepatic artery branches, represent the second cause of portal hypertension, probably of lesser significance because, even with traumatic arteriovenous aneurysm, the portal pressure does not rise to the levels seen in cirrhosis [*107*]. The third potential cause, namely distortion of vein branches by fibrosis, seems to be of less significance in the cirrhotic process.

The anastomoses between portal vein and hepatic arterial branches, by shunting blood from the parenchyma, deprive the organism of the function of the liver; just as do extrahepatic portosystemic anastomoses. Therefore, hepatic function in cirrhosis may be reduced independently of damage to liver cells in that the latter may not have adequate opportunity to act upon the constituents of the blood. Moreover, these intrahepatic vascular anastomoses, which are comparable to a large number of tiny Eck fistulas, produce a relative circulatory insufficiency of the liver which may become manifest if, as a result of other causes such as massive hemorrhage, cardiac failure or infection, the total blood flow through the liver is reduced. As a consequence there is an increased tendency for centrolobular and centronodular liver cell damage of hypoxic character. This type of liver damage, being independent of the original cause of cirrhosis and resulting from the characteristic circulatory alteration, is responsible for the tendency for progression of the cirrhotic process. Because of their parasinusoidal location, these vascular anastomoses do not relieve the portal hypertension, in contrast to the postsinusoidal obstruction of blood flow by enlarging regenerative nodules. They deserve consideration in contemplating shunt surgery because of their additive hemodynamic effects, thus explaining deterioration of hepatic function following such surgery [*108*]. Regeneratory hyperplasia after partial hepatectomy or after bile duct obstruction in the face of an existing cirrhosis produces normal liver tissue [*109,110*] possibly because the newly formed tissue does not develop such deleterious vascular anastomoses.

Ascites in cirrhosis is induced by postsinusoidal obstruction of hepatic blood flow as well as by hepatocellular damage. The interference with blood drainage not only raises the portal pressure but also, because of its postsinusoidal location, increases the formation of hepatic lymph which supposedly escapes through the hepatic capsule into the peritoneal cavity [*111, 112*]. While the contribution of hepatic lymph to ascites formation has been questioned, increase in number, dilatation and thickening of the lymphatics of the hepatoduodenal ligament has been reported in cirrhosis with ascites [*113*]. Postsinusoidal obstruction also produces congestion which in turn seems to be important in the sodium retention which rapidly develops after experimental narrowing of the inferior vena cava [*114,115*]. The site of the obstruction, in the hepatic vein tributaries, is not a specific factor since similar obstructions in other parts of the body produce a comparable type of sodium retention [*116*]. Nevertheless, together with the damage of the hepatic cells, the congestion is considered responsible for disturbed inactivation of hormones produced either by the adrenal cortex (aldosterone) [*117*] or the diencephalic-pituitary axis. Reduction of liver cell function also interferes with serum albumin formation and thus tends to reduce the osmotic pressure of the blood. In different types of cirrhosis, these factors vary in relative importance; thus in posthepatitic cirrhosis the reduction in serum albumin is considered of major significance [*118*].

CLASSIFICATION OF CIRRHOSIS

Cirrhosis can be classified according to (1) histogenesis, (2) etiology, when possible, and (3) functional characteristics [*20,106*].

Histogenetic Classification. The morphologic type of cirrhosis is best characterized by the

three pathways through which the nodules and the portohepatic septums develop [*9*].

Postnecrotic cirrhosis: Structural collapse as the result of massive, submassive or recurrent focal necrosis results in areas of scarring in which persisting sinusoids are transformed into portohepatic venous shunts and in which fragments of one or several lobules may persist as nodules. Such postnecrotic scarring, if associated with otherwise normal parenchyma (as in syphilitic hepar lobatum), does not deserve the name cirrhosis. If the necrosis develops rapidly, break fissures in the surrounding parenchyma may develop into septums with vascular anastomoses. Moreover, involvement of the surrounding parenchyma by the underlying disease leads to additional injuries, followed by formation of primary septums or nodules. Postnecrotic cirrhosis is initiated by *extensive* necrosis. Since necrotizing hepatic changes occur also in other types of cirrhosis, the designation postcollapse cirrhosis [*22*] or coarse lobular cirrhosis may be preferable. However, as in the case of the term toxic cirrhosis [*11*], these designations have not found wide acceptance. Postnecrotic cirrhosis is characterized by an uneven distribution of lesions, grossly and microscopically indicated by a wide range in the size of nodules, many with recognizable lobular architecture (Fig. 13) and broad connective tissue bands derived from primary or secondary collapse. It is often not clear whether such nodules with intact lobular architecture represent pre-existing or regenerative tissue. A third microscopic feature, the only one apparent in liver biopsy specimens, is conspicuous regeneration persisting for a long time. Any etiologic factor causing locally accentuated necrosis may produce this type of cirrhosis. It has been suggested that postnecrotic cirrhosis is characterized by a higher incidence in women, occurrence in earlier age groups and predominance of hepatic insufficiency over portal hypertension. Moreover, the nodules may be palpable in life, in contrast to other forms of cirrhosis [*10,106,119*]. Other features, like serum protein anomalies, e.g., pronounced hypergammaglobulinemia, are probably an expression of the specific etiology rather than of the histogenesis.

Diffuse septal cirrhosis: Processes involving the parenchyma throughout with equal intensity, for instance, metabolic alterations including the effects of chemical poisons, malnutrition, anoxic and congestive changes, diffuse infections, granulomas, cause diffuse development of nodules and connective tissue septums, the latter arising from aggregation of collagen membranes and traversing the lobular parenchyma. This may be associated with diffuse fatty changes. The term "diffuse septal cirrhosis" is proposed for this lesion, taking into account that under these circumstances diffuse formation of nodules of more uniform size is the rule. Although the formation of septums may be passive, they are functionally of greater significance than the active nodules. The term portal cirrhosis is frequently used for this lesion. However, this process does not necessarily start in the portal tracts [*96*]. Similarly, the use of the term Laennec's cirrhosis may create confusion [*23*]. Histologically, in the fully developed forms, hardly any portal or central fields are spared. The differentiation from focally accentuated postnecrotic cirrhosis may be especially difficult in infections, including viral hepatitis [*120*]. There is no pathognomonic clinical picture typical of diffuse septal cirrhosis.

Biliary cirrhosis: In this condition the fibrosis starts around bile ducts and ductules as a result of regurgitation of bile or of bacterial infection, or possibly as a disease *per se*. Depending upon the location of inflammation and subsequent fibrosis, either predominantly portal or intralobular periductular fibrosis is noted. The name Hanot's cirrhosis is frequently applied to the latter condition. Septums and nodules are not present and the lobular architecture is obscured but not destroyed ("avec ictère sans ascite"). Therefore, the term cirrhosis does not apply functionally to the early phases. It is justified only in the later stages, when septums and nodules form simultaneously with the development of portal hypertension and hepatocellular injury. Various types of extra- or intrahepatic cholestasis are the cause [*86*]. Clinically, hepatomegaly, icterus (with chemical evidence of cholestasis) and pruritus in the presence of relative well-being and little liver cell damage are the features common to most of them.

Frequently, several developmental pathways are represented in the same liver, creating difficulties in histogenetic classification. For instance, viral hepatitis may be the etiologic factor common to the postnecrotic, the diffuse septal and even the biliary type of cirrhosis. Moreover, diffuse septal cirrhosis may be complicated by massive necrosis with secondary collapse. The histogenetic diagnosis is even more difficult in the end stage of a shrunken, finely nodular

liver, to which the term Laennec's cirrhosis as an indication of a final common pathway best applies [*106*].

Etiologic Classification. Etiologic factors in the causation of cirrhosis are frequently listed with great assurance; however, many such statements are an oversimplification and become doubtful or untenable upon more critical survey. The evidence rests upon the history of individual patients, statistical analysis of large clinical and autopsy material, serial biopsies in individual cases, and on geographical pathology [*121*] and animal experimentation [*122*].

Malnutrition: Two main types of cirrhosis have been produced experimentally by malnutrition. One follows fatty metamorphosis induced by diets high in fat and low in choline and protein, including methionine [*10,123,124*]. A diffuse septal cirrhosis results from condensation of connective tissue capsules around fatty cysts, developing initially in the lobular center and subsequently in other areas which are relatively remote from the termination of the portal veins, such as the zones around larger portal tracts [*88,124*]. Eventually carcinoma may develop [*125*].

The other main type of cirrhosis experimentally produced by malnutrition follows the massive necrosis resulting from administration of diets deficient in cystine, vitamin E and factor 3 for several months [*10,35,127*]. The basic lesion is a disturbance of the oxidative enzyme processes [*128*] now related to an insufficient dietary intake of selenium [*129*]. After repeated attacks, the surviving animals exhibit postnecrotic cirrhosis with regenerative nodules [*10,130*].

It is very probable that human cirrhosis follows malnutrition and specifically an imbalance between intake of calories and protein, but unassailable evidence is still lacking. Besides the convincing experimental evidence [*35*], the fatty liver-cirrhosis syndrome [*131*] in man is the strongest support for this correlation. It is a time-honored clinical experience that in patients with fatty livers of long standing, particularly alcoholic persons, cirrhosis develops [*33*], different stages having been reconstructed on the basis of surveys of autopsy material [*132*] or serial liver biopsies [*38,133,134*]. However, cases have been reported of prolonged fatty metamorphosis without progression to cirrhosis [*135*]. In other instances, the transition seems to take place rapidly, justifying the term florid cirrhosis [*136*]. This suggests a link between fatty metamorphosis and cirrhosis, which seems to be hepatic necrosis to which possibly the fatty liver is more susceptible or which more readily develops in the presence of malnutrition or other factors responsible for the fatty change [*106*]. The following complicating factors are said to accelerate [*106*] the fatty liver-cirrhosis syndrome, probably by way of hepatic necrosis: (1) intercurrent infections, (2) anemia and anoxia brought on by hemorrhage, for instance, from esophageal varices, (3) cardiac failure, (4) toxic factors, (5) genetic and constitutional factors (at present poorly understood), (6) endocrine factors (greater tendency of acute aggravation in females) and (7) acute starvation. There is no evidence that viral hepatitis plays a role in this process.

The fatty liver-cirrhosis syndrome, best studied in alcoholic subjects [*15,16,131,137,138*], can be arbitrarily divided into stages (Table 1) such as simple fatty liver (massive steatosis of the French authors), fatty liver with hepatocellular degeneration, transition of fatty liver into cirrhosis, rapid transition of fatty liver into cirrhosis (florid cirrhosis) and nutritional cirrhosis with fatty change. The fatty metamorphosis may disappear at any stage of the fatty liver-cirrhosis syndrome either following correction of the nutritional imbalance or, more frequently, because of starvation since caloric undernutrition tends to remove fat from the liver [*139*].

Alcoholism: The role of alcohol in the production of cirrhosis is still not completely established. Alcohol provides excess calories and thus creates an imbalance between calories and protein or lipotropic substances such as choline or methionine. Fatty liver is produced by marginal diets supplemented by alcohol or its caloric equivalent in sucrose [*140*]. This factor of imbalance may be aggravated by the known anorexia of the alcoholic subject as well as by his tendency to gastroenteritis and pancreatitis. Moreover, alcohol is known to increase the choline requirement [*141*]. However, in addition to these factors producing imbalance in nutrition, the possibility remains that ethyl alcohol or other substances contained in alcoholic beverages exert toxic effects or, by specific depletion of enzymes, create conditioned deficiencies [*142*]. Moreover, acute alcoholism may produce circulatory disturbances in the liver. French authors stress [*137,138*] that the enlarged liver occurring in

alcoholic persons may exhibit hepatic cell degeneration without fatty change.

2. *Kwashiorkor:* Particularly in children, imbalance between calories and protein of adequate quality produces a predominantly peripheral fatty metamorphosis with stellate fibrosis associated with mental symptoms. This is found in many tropical and subtropical areas and has been designated kwashiorkor [*143*]. In Central America the term "sindrome pluricarencial de la infancia" has been used for this condition [*144*]. As a rule, the manifestations of kwashiorkor and the fatty liver disappear after the sixth year of life because of the relative reduction of requirements for lipotropic substances with deceleration of growth. Since in similar geographical areas cirrhosis, although usually without fat, is frequent in adults, the possibility has been entertained that kwashiorkor in childhood predisposes to this form of cirrhosis. However, recent experiences in various parts of the world have cast doubt on the idea that malnutrition alone is responsible for the cirrhosis occurring in adults in the tropics [*145–147*].

Gastrointestinal diseases: In various gastrointestinal disorders such as pancreatitis [*148*], ulcerative colitis [*149*] and regional ileitis [*150*], fatty liver and diffuse septal cirrhosis with or without fatty change are observed. However, this is not necessarily associated with malnutrition since other types of hepatic injury are found under these conditions [*106*].

Metabolic disorders: In galactosemia [*151*] septal cirrhosis with fatty metamorphosis is found, possibly resulting from malnutrition. The fatty liver-cirrhosis syndrome is found only rarely when malnutrition is not a contributory factor but supposedly may occur in obesity [*152*] or possibly in diabetes mellitus.

There is some suggestive evidence that a postnecrotic type of cirrhosis also may be a sequel to nutritional disturbances. For instance, in Africa this type of cirrhosis has been observed under conditions where a relation to malnutrition is possible [*55*]. Similarly, in certain metabolic disorders postnecrotic cirrhosis occasionally has been described [*151*].

Poisons: Cirrhosis, usually of the diffuse septal type, has been produced in animals by carbon tetrachloride [*154*] and various other poisons [*122*]. There is a possibility that toxic agents produce an endogenous deficiency of an essential metabolite [*155*]. This holds especially true for the cirrhosis caused by ethionine which may be either of diffuse septal or postnecrotic type [*22*].

An unquestionable history of exposure to chemical poisons without any other contributing factors is rarely elicited in cirrhotic patients. Cases following trinitrotoluene or phosphorus poisoning have been observed. As a rule cirrhosis develops only if the initial intoxication produces a massive necrosis of the character of acute yellow atrophy (as seen, for instance, following mushroom poisoning [*156*]) or if exposure to the poison persists for a long time or is repeated at short enough intervals to prevent recovery [*10,154*]. However, in most instances, exposure to poisons is followed by either death or complete recovery.

Alteration of Metabolism of Heavy Metals. In both hemochromatosis and hepatolenticular degeneration (Wilson's disease), excessive accumulation of metals occurs. However, these deposits do not appear to be the direct causes of the hepatic injury.

Hemochromatosis: In idiopathic hemochromatosis, which is associated with a diffuse septal cirrhosis characterized by relatively thick connective tissue septums, an excess of granular iron pigment is noted within the polygonal cells and ductular cells of the liver as well as extracellularly, primarily in the portal tracts. Relatively little iron is found within Kupffer's cells in which it aggregates only where liver cells degenerate. This distribution, which suggests an alteration of the cellular iron metabolism and which is associated also with the deposition of iron-free hemofuscin pigment, is different from that seen in hemosiderosis in which the iron is found mainly in the reticuloendothelial cells, apparently as the result of excessive availability of hemoglobin derived, for instance, from blood transfusions or intravascular hemolysis. The assumption of a basic difference [*106,157,158*] between hemochromatosis as an essential parenchymal siderosis with functional alterations of the organs involved and hemosiderosis as reticuloendothelial iron deposition without functional disturbance is not universally accepted. Some investigators assume only differences in degree between these two conditions [*159*]. In recent years hemochromatosis, with the classical clinical manifestations of parenchymal organ involvement including hepatic insufficiency, diabetes, cardiac failure and endocrine disturbances, has been reported in patients suffering from chronic anemia most of whom received nu-

merous blood transfusions. This has been designated secondary hemochromatosis [*160,161*], exogenous hemochromatosis [*162*] or transfusional siderosis [*163*], although it may be impossible in some of these reported cases to assign them clearly either to parenchymal or reticuloendothelial siderosis. There is no question that in a number of patients with chronic anemias the manifestations of a parenchymal siderosis (hemochromatosis) develop. In contrast to idiopathic hemochromatosis, which requires several decades for development, this form of hemochromatosis produces clinical manifestations in a matter of years. In this florid hemochromatosis [*164*] a preponderance of male patients, which is typical of primary hemochromatosis, is not observed. Iron overload from blood transfusions is not the causative but only a contributing factor since cases have been reported after few blood transfusions [*161*] and in some instances the hepatic iron content by far exceeds the amount of transfused iron [*165*]. It appears rather that proliferation of cells of the erythropoietic series, as occurs in erythroid myelosis, may represent an important factor in the apparent increased iron avidity of the parenchymal liver cells [*164*]. This raises the question whether or not the parenchymal cell iron deposits are responsible for the changes observed, including the cirrhosis. In florid hemochromatosis periportal accumulation of ductular cells heavily loaded with iron is associated with the appearance of excessive reticulum fibers, and with only gradual and less marked accumulation of collagenous membranes. Since the iron is deposited in the mitochondria [*166*], presumably damaging them, this type of fibrosis possibly is comparable to that associated with hepatocellular degeneration and transformation into ductular cells [*167*]. These morphologic observations suggest that excessive parenchymal iron deposition, whether brought about by an inborn error of iron metabolism or by altered utilization of iron by the red cells, is the principal cause of this type of cirrhosis. Further investigation will be necessary to exclude the possibility that iron accumulation and cirrhosis are independent results of the same cause or that the liver loaded with iron becomes susceptible to the effects of other injurious agents, as is assumed to be the case in the fatty liver-cirrhosis syndrome.

Hepatolenticular degeneration: The copper deposition visualized in the hepatic cells [*168*] has been associated with a congenital and familial defect of the copper-carrying serum protein, ceruloplasmin [*169,170*]. Morphologically, the picture usually is that of a postnecrotic cirrhosis [*151*] which in acute cases exhibits marked activity and conspicuous liver cell destruction. Since the lesions in this type of cirrhosis are not uniformly distributed throughout the liver, it is not easily correlated with diffuse copper deposition and it therefore appears possible that other factors play a role, such as increased susceptibility to viral infections or nutritional disturbances of endogenous nature.

Cholestasis: The role of cholestasis in producing cirrhosis is better substantiated than that of many other etiologic factors. Non-infected extrahepatic biliary obstruction produces fully developed cirrhosis as a rule only in infants with congenital malformations of the biliary tract since adults usually do not survive the obstruction long enough. In adults, secondary biliary cirrhosis (associated with extrahepatic obstruction) develops usually with incomplete obstruction and infection, such as is usually found after strictures of the common duct. This form of cholangitic cirrhosis [*86,106*] therefore has a twofold histogenesis, namely, biliary stasis and portal inflammation. Primary biliary cirrhosis results from prolonged intrahepatic cholestasis [*171*]. However, the mechanism is conjectural. Only in the late stages are there functional manifestations of cirrhosis. The disease may be associated with hypercholesteremia and cutaneous xanthomatosis which are considered secondary to the hepatic lesion. Nevertheless, pruritus and hypercholesteremia sometimes precede the jaundice. The causes of the intrahepatic cholestasis or cholangiolitis are still argued [*172*]. The high incidence in women at the end of the reproductive period suggests an endocrine factor. The occurrence in young soldiers suggests viral hepatitis. Whether or not the cholestasis following intake of drugs such as chlorpromazine or methyltestosterone leads to cirrhosis remains to be proved.

Anoxia and congestion: Interference with the drainage of blood from the liver, as seen in long-standing cardiac failure, produces centrolobular fibrosis because of mechanical pressure upon the liver cells, together with anoxia. Only if the process becomes very severe, as in tricuspid insufficiency or constrictive pericarditis, does the dissection of the lobular pattern by septums reaching from the center to the portal

tracts become extensive enough to justify the term cardiac cirrhosis. This type of cirrhosis as a rule is septal and small-nodular but otherwise shows an irregular distribution. Prolonged sinusoidal stagnation with gradual vascular occlusion, as in sickle cell anemia, occasionally results in a postnecrotic type of cirrhosis [*173*]. Rh incompatibility in infants also has been associated with cirrhosis [*174*]. Cirrhotic transformation in the presence of and probably secondary to widespread carcinoma of the liver is probably a reflection of disturbance of hepatic blood flow.

Diffuse hepatic granulomatosis: Various granulomatous disorders such as brucellosis [*175*] sarcoidosis, tuberculosis and histoplasmosis [*10*], also parasitic diseases, have been considered to be causes of diffuse septal cirrhosis. However, in most instances only the initial stages of septum formation extending from the portal tracts and a few regenerative nodules are noted.

Viral hepatitis: There is convincing evidence that massive necrotic viral hepatitis progresses to postnecrotic cirrhosis [*120,176*]. If viral hepatitis were a common cause of cirrhosis, the spotty necrotic type which is far more frequent than the massive necrotic type would have to be a common antecedent of cirrhosis. This raises the question whether diffuse septal or Laennec's cirrhosis may be included in the category of posthepatitic cirrhosis [*120,177,178*]. Liver biopsy specimens in the subacute stage of spotty necrotic viral hepatitis indicate beginning septum formation starting usually from the portal tracts. The transition of active viral hepatitis into diffuse septal cirrhosis, however, is not well documented by serial biopsy as yet. A survey of soldiers with viral hepatitis showed a surprisingly low incidence of cirrhosis [*179*]. This may not necessarily hold true for civilians in whom a higher incidence was noted [*180,181*]. The probability of great geographical difference throughout the world deserves emphasis. The possibility that viral hepatitis may lead to primary biliary cirrhosis has been referred to; thus posthepatitic cirrhosis may show the landmarks of all three histogenetic pathways.

Unknown etiology: Typical postnecrotic cirrhosis [*119*] is observed more commonly among women and as a rule shows a high thymol turbidity, elevation of serum gamma globulin above 3 gm. per 100 ml. and distinctly reduced serum mucoproteins without any antecedent history suggesting massive necrosis of the liver. Alcoholism is not frequent and a history of exposure to poisonous agents cannot be elicited. Clinically, silent viral hepatitis without jaundice cannot be excluded in view of the fact that in rats a similar lesion develops from recurring focal necrosis without jaundice or other obvious indication. The frequent occurrence of this type of cirrhosis in African natives may be the result of viral hepatitis or malnutrition. However, it appears wise at present to recognize our lack of precise information about the etiology of many instances of postnecrotic cirrhosis in order not to discourage continued search for etiologic factors. For instance, the postnecrotic type of cirrhosis occurring in young women may possibly be the result of a hypersensitivity to estrogens [*182*]. A diffuse septal cirrhosis of unknown etiology is encountered in various parts of the world, including South America. Also, the etiology of cirrhosis in infants and children, although frequently suggested to be of viral origin and usually characterized by cholestasis and multinucleated liver giant cells, requires further elucidation [*183–185*].

Combination of etiologic factors: In a large number of cases of cirrhosis the synergistic action of several etiologic factors best explains the morphologic picture. (1) *Congestion and malnutrition.* While passive congestion may not be of sufficient degree to produce cirrhosis, when combined with malnutrition and/or alcoholism cirrhosis very frequently results [*186*]. (2) *Tuberculosis and malnutrition.* Similarly in tuberculosis, the association with alcoholism facilitates the development of a diffuse septal cirrhosis [*187*]. (3) *Schistosomiasis.* There seems to be gradual agreement, at least in South America [*188*] and Egypt [*189*], that schistosomiasis leads to extensive portal fibrosis (lesion of Symmers or "pipe stem cirrhosis") associated with portal hypertension but without destruction of the lobular architecture. When diffuse septal cirrhosis is found with schistosomiasis, it is probably not the result of toxic factors derived from the parasite but a consequence of the associated malnutrition. (4) *Fibrocystic disease of the pancreas.* With the present improved treatment, patients with mucoviscidosis have an increased life span. With this prolonged duration hepatic sequelae have become apparent, the result of such factors as focal intrahepatic biliary obstruction owing to mucoviscidosis and its concomitant malnutrition, and possibly also of infection. Postnecrotic cirrhosis has been observed [*190*] as well as focal

biliary cirrhosis [*191*], also with fatty change [*192*]. (5) *Venoocclusive disease.* Malnourished Central American children have a fatal form of cirrhosis which results from endophlebitic thickening and eventual obstruction of hepatic vein branches [*193*]. The symptoms of Budd-Chiari's disease eventually develop, with severe ascites but usually without jaundice. Evidence is accumulating that the senecio and crotalaria alkaloids from plants used to prepare a beverage ("bush tea") are the cause of this syndrome when acting together with malnutrition [*194*]. Similar lesions have been found in man and in animals in South Africa where related poisonous plants grow in pastures [*195*].

Conditions Resembling Cirrhosis. Conspicuous disfiguration of the liver, sometimes with extensive fibrosis, may occur without functional manifestations of cirrhosis (with the exception of portal hypertension) if the lobular architecture is preserved in the greater part of the organ.

Hepatic fibrosis resulting from subsided hepatic congestion: At autopsy there is occasionally observed a finely granular liver which on cut surface exhibits loss of the normal lobular arrangement and the suggestion of a nodular architecture in the absence of any clinical or functional manifestations of liver disease. Microscopically there is an irregular, partly centrolobular and partly portal fibrosis, without portohepatic septums or regenerative nodules. In such instances the history or the anatomic findings in organs other than the liver bear witness to previous episodes of cardiac failure during which the architecture of the liver was apparently distorted but not destroyed.

Gaucher's disease: In adults with reticuloendothelial cerebrosidosis of Gaucher, a diffuse thickening of Glisson's capsule of the much enlarged liver is noted, with extension of thick connective tissue bands into the parenchyma. In addition, an irregular connective tissue scaffolding traverses the interior of the liver, involving the central zone and surrounding accumulations of Gaucher's cells. Nevertheless, the basic architecture is preserved and manifestations referable to hepatic failure do not belong to the picture of Gaucher's disease [*151*].

Syphilitic hepar lobatum: Although the hepatic enlargement in tertiary syphilis may be associated with marked disfiguration of the enlarged organ and with portal hypertension, the lobular architecture is histologically preserved except for areas of collapse.

Hamartomas: The lobular architecture is sometimes altered in circumscribed areas of the liver, particularly in the vicinity of post-traumatic or space-occupying lesions or as a result of a tumor-like focal congenital defect in the organization of the tissue. This may take the form of focal cirrhosis [*196*] or of multiple cystic ductular proliferation in the portal tracts surrounded by a dense mass of collagenous fibers (v. Meyenburg complexes). Portal hypertension may be present [*197,198*]. Telangiectatic cirrhosis in Osler's disease also causes portal hypertension [*199*].

Functional Classification. Apart from morphogenetic and etiologic connotations, cirrhosis may be described in terms of functional characteristics, depending upon the presence and degree of (1) hepatic failure, (2) portal hypertension, (3) jaundice, (4) extent and (5) activity of the cirrhotic process [*3,20*]. These criteria indicate much more than histogenesis and etiology the clinical status of the patient, prognosis, and the medical and surgical therapy to be employed.

Hepatic failure: This feature, as evidenced by clinical and laboratory manifestations, may result from different processes: (1) Hepatocellular insufficiency resulting from degeneration or necrosis of the liver cells is due to impaired function of the hepatic parenchyma and is readily reflected in alterations in liver biopsy specimens which in turn mirror the results of hepatic tests. (2) Hepatocirculatory insufficiency resulting from shunt of blood bypassing the hepatic parenchyma either by portohepatic venous anastomoses within the liver or portosystemic anastomoses outside the liver does not correlate well with the laboratory manifestations of hepatic failure; it may occur without jaundice and without significant histological damage in biopsy or autopsy specimens. (3) Severe prolonged cholestasis may be associated with fatal hepatic failure although at autopsy, except for terminal centrolobular necrosis, only the evidence of cholestasis is seen; here the cause for hepatic failure is not clear. (4) In huge fatty livers, sometimes with beginning cirrhosis, excessive fatty metamorphosis may be associated with relatively little if any necrosis, although the patient dies in hepatic failure. The cause of this is not understood [*16*]. The coma caused by excess ammonia (ammoniacal encephalopathy [*106*], implies hepatocirculatory rather than hepatocellular hepatic failure in that ammonia

formed outside the liver, especially in the intestine, reaches the brain without being metabolized by the liver (portosystemic encephalopathy) [*200*]. Since hepatic coma in cirrhosis is apparently more often the result of circulatory than hepatic failure [*201*], it is more readily reversible than the coma of acute hepatitis due to destruction of liver cells.

Portal hypertension: The portal pressure is raised not only in cirrhosis but also in hepatitis and fatty liver without cirrhotic manifestations. However, in the last two conditions it falls after appropriate therapy [*202*]. Portal hypertension produces splenomegaly and venous collaterals, of which esophageal varices are the most important. It is not established, however, whether or not portal hypertension is the only causative factor. Large varices may exist in the absence of portal hypertension [*89*] and their size may vary in the course of persistent portal hypertension [*203*]. It is therefore entirely possible that endocrine factors such as those implicated in cutaneous spider formation may play a role in the genesis of varices [*204*]. But irrespective of pathogenesis, bleeding esophageal varices together with hepatic failure account for the vast majority of fatalities in cirrhotic subjects [*205,206*].

Jaundice: Liver cell damage in cirrhosis is often associated with icterus. However, the degree of jaundice frequently is out of proportion to the degree of liver cell injury [*19*] and bile pigment is found not only within the liver cells, staining coagulated protein clumps of varying sizes, but also in bile canaliculi in the form of plugs and even as microcalculi in the septal bile ducts. This clearly indicates that the bile pigment has passed through the liver cells. Therefore, either stasis or overproduction of bile (because of hemolysis) accounts for the jaundice in cirrhosis, rather than liver cell damage. A hemolytic component is reflected in the shortened life span of red cells in cirrhosis [*207*]. However, since much of the serum bilirubin is direct reacting and glucuronic acid conjugation of the bilirubin is intact, bile stasis has to be postulated as the most important factor. The intrahepatic cholestasis reflected in increased serum alkaline phosphatase activity and total cholesterol levels cannot be explained by obstruction or compression of intrahepatic bile ducts since the latter is seldom widespread enough to prevent the non-involved bile ducts from compensatory excretion. Obstruction of one main hepatic duct causes elevation of the serum alkaline phosphatase but not necessarily of bilirubin [*208*]. These considerations led to the assumption that the sites of involvement in intrahepatic cholestasis are the ductules. This gave rise to the term, cholangiolitis [*209*]. Subsequent studies of patients with acute intrahepatic cholestasis failed to indicate any inflammation in the portal tracts [*85*]. This appears only in the subacute stages or as transient portal inflammatory exudate with admixture of eosinophils after the use of such drugs as arsenicals, chlorpromazine, thiouracil and para-aminosalicylic acid. This cellular infiltrate may disappear although the cholestasis progresses to fatality or it may be conspicuous in the absence of jaundice, as seen in serial biopsy studies [*210*]. The obscurities of the histologic picture of acute intrahepatic cholestasis can best be reconciled by postulating a change in the membrane either of the canaliculi or of the ductules. Altered permeability might permit either escape of protein into the bile or its regurgitation or reabsorption of biliary solutes with inspissation of bile. The microvilli forming the luminal membrane of liver cells and bile ductules, as seen with the electron microscope, may be the site of this membrane effect. Inspissation produces bile casts and microcalculi which in turn add an obstructive component. Regurgitation of bile may account for periductular inflammation and subsequent fibrosis; the bile ductules may be compressed thereby or even choked off. This produces secondary mechanical obstruction for which adequate morphologic evidence exists [*86*]. Intrahepatic cholestasis may accompany hepatic disorders in which liver cell damage is in the foreground, such as postnecrotic cirrhosis, viral hepatitis [*211*] or fatty liver-cirrhosis syndrome [*212*], and under these circumstances may create diagnostic difficulties (cholestatic phase of cirrhosis). It thus appears that intrahepatic cholestasis represents a non-specific response of the liver to a variety of insults. The nosologic mechanism is discussed especially by French authors [*213*]. They have recognized three varieties of chronic cholestasis [*214*]: (1) obstructive intrahepatic cholangiolitis of Roessle complicating extrahepatic cholangitis and occurring in both sexes, (2) pericholangiolitis of MacMahon destroying the septal bile ducts and occurring in women with or without xanthomatosis, and (3) diffuse mesenchymatous hepatitis with nodular lymphomatosis of Hanot and Kiener

TABLE I

STAGES IN THE FATTY LIVER-CIRRHOSIS SYNDROME IN ALCHOLIC SUBJECTS AS OBSERVED AT AUTOPSY [*16,132,136*]

Diagnosis	Approx. Predominant Age (yr.)	% Males	Hepatomegaly	Splenomegaly	Ascites	Esophageal Varices	Results of Hepatic Tests
Simple fatty liver	50	70	++	+	±	0	Not necessarily abnormal
Fatty liver with hepatocellular failure and jaundice	37	45	++++	+	+	+	Cephalin flocculation and thymol turbidity tests abnormal in more than 40%; cholestasis in 30%
Florid cirrhosis	40	63	++	++	++	+++	Thymol turbidity and especially cephalin flocculation tests almost always abnormal; cholestasis in over 30%
Septal cirrhosis without hepatic failure	53	63	+++	++	+++	++	Variable results; thymol turbidity usually normal
Septal cirrhosis with hepatic failure	48	50	+++	+++	++++	++++	Cephalin flocculation in 70%; thymol turbidity in 30% abnormal; cholestasis in almost half of cases

occurring in men and destroying the intra- and perilobular ductules and supposedly responsive to steroid therapy.

Extent of cirrhosis formation: The extent of the cirrhotic process depends on the degree to which the original lobular parenchyma is replaced by regenerative nodules and on the number of septal connections between portal triads and central fields. Few clinical and laboratory manifestations indicate the difference between minimal, moderately advanced and far advanced cirrhosis [*20*]. Splenomegaly and edema are more frequent in the far advanced stage. Cirrhosis in this stage has been recognized incidentally at autopsy or on biopsy in the absence of clinical or laboratory manifestations (latent cirrhosis) [*215*].

Activity of cirrhosis: In contrast to the extent of cirrhosis, the rate of progression of cirrhotic changes is of major prognostic and therapeutic significance. Cases of progressive cirrhosis may be differentiated from those of arrested cirrhosis, upon statistical analysis, by increased incidence of jaundice, splenomegaly, spider nevi and palmar erythema [*20*]. Progression is also indicated by an increase in the titer of thymol turbidity and cephalin flocculation tests, a rising serum gamma globulin and falling serum albumin. In biopsy and autopsy specimens, the rate of progression is mirrored in (1) active regeneration, (2) extension of increased reticulum fibers and collagenous membranes throughout the parenchyma with beginning formation of septums, (3) inflammatory cells around areas of parenchymal necrosis as well as in septums and portal triads, and (4) indistinct boundaries between parenchyma and portal tracts or septums with destruction of the limiting plate and excessive proliferation of ductular cells. The term florid cirrhosis is used to indicate a condition in which the extent of the cirrhotic process is still minimal while rapid progression takes place. Such florid forms in the fatty liver-cirrhosis syndrome [*136*] are readily mistaken for an acute or subacute hepatitis on clinical grounds, in contrast to far advanced but arrested forms of cirrhosis.

SUMMARY

Cirrhosis is characterized by interdependent parenchymal and mesenchymal processes. Fibrosis in cirrhosis may be the result of primary "irritation" of the connective tissue or of increased pressure in the hepatic vein tributaries (law of Thomas), or it may be stimulated by epithelial alterations such as hepatocellular necrosis with subsequent stromal collapse, de-

generation or fatty metamorphosis of liver cells, and excess of ductular cells. Anatomically, cirrhosis is defined by an altered reconstruction of the lobular architecture reflected in regenerative nodules and in connective tissue septums linking portal canals and centrolobular fields and carrying vascular anastomoses. The formation of nodules and septums is dovetailed; either may be active or passive in relation to the other.

Cirrhosis is best classified according to (1) histogenesis, (2) etiologic factors, and (3) functional alterations. The histogenetic classification depends upon the three pathways through which the architecture is reconstructed (postnecrotic, diffuse septal and biliary). The etiological classification is hampered by insufficient knowledge of the etiological causes of the several forms of cirrhosis. The functional characteristics of cirrhosis include hepatic failure (either hepatocellular or hepatocirculatory), portal hypertension, cholestasis, extent and activity of the cirrhotic process. In respect to prognosis and therapy, the extent of cirrhosis is of less significance than the activity of the cirrhotic lesion.

REFERENCES

1. Proceedings of the International Society of Geographic Pathology, Geneva, 1931.
2. Goldblatt, H. In: Liver Injury. Transactions of the Sixth Conference, May 1–2, 1947. New York, 1947. Josiah Macy, Jr. Foundation.
3. Fifth Panamerican Congress of Gastroenterology. Havana, Cuba, 1956. Report of the Board for classification and nomenclature of cirrhosis of the liver.
4. Cirrhosis of the Liver: the Leading Cause of Death. New York, 1957. Metropolitan Life Insurance Co. Statistical Bulletin.
5. Moon, V. H. Experimental cirrhosis in relation to human cirrhosis. *Arch. Path.*, 18: 381, 1934.
6. Karsner, H. T. Morphology and pathogenesis of hepatic cirrhosis. *Am. J. Clin. Path.*, 13: 569, 1943.
7. Baggenstoss, A. H. The significance of nodular regeneration in cirrhosis of the liver. *Am. J. Clin. Path.*, 25: 936, 1955.
8. Fiessinger, N. and Walter, H. Nouveaux Procedes D'Exploration Fonctionelle du Foie. Paris. 1934. Masson et Cie.
9. Popper, H. and Elias, H. Histogenesis of hepatic cirrhosis studied by the three-dimensional approach. *Am. J. Path.*, 31: 405, 1955.
10. Himsworth, H. P. Lectures on the Liver and Its Diseases. Cambridge, 1947. Harvard University Press.
11. Mallory, F. B. Cirrhosis of liver (Shattuck lecture). *New England J. Med.*, 206: 1231, 1932.
12. Roessle, R. In: Henke, F. and Lubarsch, O. (editors). Handbuch der speziellen pathologischen Anatomie und Histologie, vol. 5, pt. 1, p. 243. Berlin, 1930. Springer.
13. Smetana, H. Pathologic anatomy of early stages of viral hepatitis. In: Hepatitis Frontiers, pp. 77–111. Boston, 1956. Little, Brown & Co.
14. Koch-Weser, D., Huerga, J. de la, Yesinick, C. and Popper, H. Hepatic necrosis due to bromobenzene as an example of conditioned amino acid deficiency. *Metabolism*, 2: 248, 1953.
15. Davidson, C. S. and Phillips, G. B. Acute hepatic insufficiency in the chronic alcoholic. Clinical and pathological study. *Arch. Int. Med.*, 94: 585, 1954.
16. Popper, H. and Szanto, P. B. Fatty liver with hepatic failure in alcoholics. *J. Mt. Sinai Hosp.*, 24: 1121, 1957.
17. Altman, H. W. Über Leberveränderungen bei allgemeinem Sauerstoffmangel nach Unterdruckexperimenten an Katzen. *Frankfurt. Ztschr. Path.*, 60: 376, 1949.
18. Dubin, I. N. Anoxic necrosis of liver in man. *Am. J. Path.*, 33: 589, 1957.
19. Waldstein, S. S., Popper, H., Szanto, P. B. and Steigmann, F. Liver cirrhosis: relation between function and structure based on biopsy studies. *Arch. Int. Med.*, 87: 844, 1951.
20. Schaffner, F., Popper, H. and Dalla Torre, M. Structural alterations in the clinical evaluation of cirrhosis. *Gastroenterology*, 30: 357, 1956.
21. Welin, G. Needle biopsy and liver function in earlier stages of fatty cirrhosis of the liver. *Acta med. Scandinav.* (supp. 246), p. 260, 1950.
22. Popper, H., Clearkin, K., Kent, G., Parthasarathy, M. and Bruce, C. Coarse nodular cirrhosis produced by ethionine. *Am. J. Path.*, 31: 594, 1955.
23. Popper, H. Liver disease—morphologic considerations. *Am. J. Med.*, 16: 98, 1954.
24. Ricketts, W. E., Kirsner, J. B., Palmer, W. L. and Sterling, K. Observations on the diagnostic value of liver biopsy, tests of hepatic function and fractionation of serum proteins in asymptomatic portal cirrhosis. *J. Lab. & Clin. Med.*, 35: 403, 1950.
25. Post, J. and Rose, J. V. Clinical, functional and histologic studies in Laënnec's cirrhosis of the liver. *Am. J. Med.*, 8: 300, 1950.
26. Shils, M. E., DeGiovanni, R. and Stewart, W. B. Fatty liver of portal type: effect of choline, methionine and vitamin B_{12}. *J. Nutrition*, 56: 95, 1955.
27. Greenspan, E. and Dreiling, D. Serum mucoprotein level in differentiation of hepatogenic from obstructive jaundice. *Arch. Int. Med.*, 91: 474, 1953.
28. Zinke, M. R. and Leevy, C. M. Correlation of serum transaminase with histopathology in Laënnec's cirrhosis. *Gastroenterology.* (In press.)
29. Nino-Herrera, H., Harper, A. E. and Elvehjem, C. A. Histological differentiation of fatty livers produced by threonine or choline deficiency. *J. Nutrition*, 53: 469, 1954.
30. Whiteside, M. G. The development of diffuse nutritional fibrosis in the liver of rats. *Australasian Ann. Med.*, 5: 294, 1956.

31. Hartroft, W. S. Periportal fatty casts and reticulin condensation in livers of rats subjected to cyclic protein feeding. *Federation Proc.*, 16: 359, 1957.
32. Follis, R. H., Jr. A Kwashiorkor-like syndrome observed in monkeys fed maize. *Proc. Soc. Exper. Biol. & Med.*, 96: 523, 1957.
33. Connor, C. L. The etiology and pathogenesis of alcoholic cirrhosis of the liver. *J. A. M. A.*, 112: 387, 1939.
34. Davies, J. N. P. Nutrition and nutritional disease. *Am. Rev. Med.*, 3: 99, 1952.
35. Gyorgy, P. Experimental hepatic injury. *Am. J. Digest. Dis.*, 19: 392, 1952.
36. Dible, J. H. Degeneration, necrosis and fibrosis in the liver. *Brit. M. J.*, 1: 833, 1951.
37. Elias, H. and Popper, H. Venous distribution in livers. *Arch. Path.*, 59: 33, 1955.
38. Buck, R. E. Observations on alcoholic fatty liver; use of interval needle biopsy and liver function tests. *J. Lab. & Clin. Med.*, 33: 555, 1948.
39. Arata, D., Svenneby, G., Williams, J. N., Jr. and Elvehjem, C. A. Metabolic factors and the development of fatty livers in partial threonine deficiency. *J. Biol. Chem.*, 219: 327, 1956.
40. Kent, G., Popper, H., Dubin, A. and Bruce, C. The spleen in ethionine induced cirrhosis. Its role in γ-globulin elevation. *Arch. Path.*, 64: 398, 1957.
41. Briellmann, A. Über Plasmazellenbefunde in Lymphknoten bei Lebercirrhosen und die Bedeutung der Russelschen Körperchen. *Schweiz. Ztschr. allg. Path.*, 18: 23, 1955.
42. Glagov, S., Kent, G. and Popper, H. In preparation.
43. Mackay, J. R., Taft, L. I. and Cowling, D. C. Lupoid hepatitis. *Lancet*, 2: 1323, 1956.
44. Gajdusek, D. C. and MacKay, I. R. An "auto-immune" reaction against human tissue antigens in certain acute and chronic diseases. *Arch. Int. Med.*, 101: 9, 1958.
45. Moschcowitz, E. The pathogenesis of splenomegaly in hypertension of the portal circulation. Congestive splenomegaly. *Medicine*, 27: 187, 1948.
46. Bucher, N. L. R., Scott, J. F. and Aub, J. C. Regeneration of the liver in parabiotic rats. *Cancer Res.*, 11: 457, 1951.
47. Glinos, A. D. and Gey, G. O. Humoral factors in the induction of liver regeneration in the rat. *Proc. Soc. Exper. Biol. & Med.* 80: 421, 1952.
48. Blomquist, K. Growth stimulation in the liver and tumour development following intraperitoneal injections of liver homogenates in the rat. *Acta path. et microbiol. Scandinav.*, supp. 121, 1957.
49. Himes, M., Hoffman, J., Pollister, A. W. and Post, J. Origin of polyploid nuclei in rat livers during regeneration following carbon tetrachloride poisoning. *J. Mt. Sinai Hosp.*, 24: 935, 1957.
50. Craig, J. M. and Landing, B. H. Form of hepatitis in neonatal period simulating biliary atresia. *Arch. Path.*, 54: 321, 1952.
51. Kent, G. Popper, H. and Bruce, C. Hepatic lesions induced by ethionine. To be published.
52. Smetana, H. Histogenesis of coarse nodular cirrhosis. *Lab. Invest.*, 5: 175, 1956.
53. Edmondson, H. A. and Steiner, P. Primary carcinoma of the liver. *Cancer*, 7: 462, 1954.
54. Köhne, K. Über das Vorkommen von Mikrocarcinomen in cirrhotischen Lebern. *Ztschr. Krebsforsch.*, 61: 350, 1956.
55. Higginson, J., Grobbelaar, M. B. and Walker, A. R. P. Hepatic fibrosis and cirrhosis in man in relation to malnutrition. *Am. J. Path.*, 33: 29, 1957.
56. MacDonald, R. A. Cirrhosis and primary carcinoma of the liver. *New England J. Med.*, 255: 1179, 1956.
57. Roth, D. and Duncan, P. A. Primary carcinoma of the liver after giant-cell hepatitis of infancy. *Cancer*, 8: 986, 1955.
58. Cachera, R. and Darnis, F. Examen du foie humain (preleve par ponctionbiopsie) au microscope electronique. *Rev. franç. d'études Clin. Biol.*, 1: 296, 1956.
59. Horstmann, E. Entwicklungsbedingungen des intrahepatischen Gallengangsystems. *Arch. f. Entwecklngsmech. d. Organ.*, 139: 363, 1939.
60. Farber, E. Similarities in the sequence of early histological changes induced in the liver of the rat by ethionine, 2-acetylaminofluorene and 3'-methyl-4-dimethyaminoazobenzene. *Cancer Res.*, 16: 142, 1956.
61. Gillman, J., Gilbert, C. and Spence, I. Some factors regulating the structural integrity of the intrahepatic bile ducts with special reference to primary carcinoma of the liver and vitamin A. *Cancer*, 7: 1109, 1954.
62. Lopez, M. Proliferazioni iperplastiche e neoplastiche delle vie bilifere intrahepatiche da tioacetamide. *Experientia*, 12: 185, 1956.
63. Schoental, R. and Magee, P. N. Chronic liver changes in rats after a single dose of lasiocarpine, a pyrrolizidine (senecio) alkaloid. *J. Path. Bact.*, 74: 305, 1957.
64. Popper, H., Kent, G. and Stein, R. Ductular cell reaction in the liver in hepatic injury. *J. Mt. Sinai Hosp.*, 24: 551, 1957.
65. Elias, H. De morphologia carcinomatis primarii hepatis humani et de eius contextu cum evolutione phylogenetica et ontogenetica. *Acta Hepatol.*, 5: 1, 1957.
66. Stein, R., Kent, G. and Popper, H. In preparation.
67. Gersh, I. Ground substance and the plasticity of connective tissues. *Harvey Lect.*, pp. 211–241, 1949–1950.
68. Elias, H. and Spanier, S. H. Structure of the collagenous tissue in the cirrhotic liver, a contribution to the geometry of sectioning. *Ztschr. f. wissensch. Mikr.*, 61: 312, 1953.
69. Brewer, D. B. Differences in the fine structure of collagen and reticulin as revealed by the polarising microscope. *J. Path. Bact.*, 74: 371, 1957.
70. Kramer, H. and Little, K. Nature of reticulin. *Nature, London*, 170: 499, 1952.
71. Asboe-Hansen, G. Editor. Connective Tissue in Health and Disease. Copenhagen, 1954. Ejnar Munksgaard.
72. Kent, G., Dubin, A., Fels, I. G. and Popper, H. In preparation.
73. Windrum, G. M., Kent, P. W. and Eastoe, J. E. The constitution of human renal reticulum. *Brit. J. Exper. Path.*, 36: 49, 1955.

74. Leblond, C. P., Glegg, R. E. and Eldinger, D. Presence of carbohydrates with free 1, 2 glycol groups in sites stained by the periodic-acid-Schiff technique. *J. Histochem.*, 5: 445, 1957.
75. Robb-Smith, A. H. T. The reticulin riddle. *J. Mt. Sinai Hosp.*, 24: 1155, 1957.
76. Morrione, T. G. Factors influencing collagen content in experimental cirrhosis. *Am. J. Path.*, 25: 273, 1949.
77. Balasubrahmanyan, M. The ground substance of the fibrous tissue in experimental liver cirrhosis. *J. Path. Bact.*, 65: 123, 1953.
78. Cameron, G. R. and Hassan, S. M. Repair of Glisson's capsule after tangential wounds of the liver. *J. Path. Bact.*, 73: 1, 1957.
79. Ungar, H. and Feldman, J. D. The absorption of collagen in the liver, induced by the implantation of plain surgical gut. *Am. J. Path.*, 29: 463, 1953.
80. Williams, G. A histological study of the connective tissue reaction to carrageenin. *J. Path. Bact.*, 73: 557, 1957.
81. Zak, F. G., Bubelis, I., Paronetto, F. and Miller, P. Unpublished observations.
82. McDonald, R. A. Personal communication.
83. Hartroft, W. S. and Ridout, J. H. Pathogenesis of the cirrhosis produced by choline deficiency. *Am. J. Path.*, 27: 951, 1951.
84. Urteaga Ballon, O. Contribucion a la histopatologia de las cirrosis y seudo-cirrosis hepaticas. La elastosis como substratum anatomo-patologico de la cirrosis de Laënnec. *Arch. peruanos pat. y clín.*, 2: 3, 1948.
85. Popper, H. and Szanto, P. B. Intrahepatic cholestasis ("cholangiolitis"). *Gastroenterology*, 31: 683, 1956.
86. MacMahon, H. E. Biliary cirrhosis. Differential features of the five types. *Lab. Invest.*, 4: 243, 1955.
87. Popper, H., Szanto, P. B. and Elias, H. Transition of fatty liver into cirrhosis. *Gastroenterology*, 28: 183, 1955.
88. Rappaport, A. M. Anatomic Considerations in Disease of the Liver. Edited by Schiff, L. Philadelphia, 1956. J. B. Lippincott Co.
89. Turner, M. D., Sherlock, S. and Steiner, R. E. Splenic venography and intrasplenic pressure measurement in the clinical investigation of the portal venous system. *Am. J. Med.*, 23: 846, 1957.
90. Madden, J. L., Lore, J. M., Jr., Gerold, F. P. and Ravid, J. Intrahepatic circulation in normal and in diseased liver. *S. Forum*, 4: 380, 1953.
91. Kelty, R. H., Baggenstoss, A. H. and Butt, H. R. Relation of regenerated liver nodule to vascular bed in cirrhosis. *Gastroenterology*, 15: 285, 1950.
92. Popper, H., Elias, H. and Petty, D. E. Vascular patterns of the cirrhotic liver. *Am. J. Clin. Path.*, 22: 717, 1952.
93. Hales, M. R., Allan, J. S. and Hall, E. M. Injection corrosion studies of the vascular systems of normal and cirrhotic livers. *Am. J. Path.*, 31: 595, 1955.
94. Baggenstoss, A. H. and Stauffer, M. H. Posthepatic and alcoholic cirrhosis: clinicopathologic study of 43 cases of each. *Gastroenterology*, 22: 157, 1952.
95. Breedis, C. and Young, G. The blood supply of neoplasms in the liver. *Am. J. Path.*, 30, 969, 1954.
96. Ashburn, L. L., Endicott, K. M., Daft, F. S. and Lillie, R. D. The nonportal distribution of the trabeculae in dietary cirrhosis of rats and in carbon tetrachloride cirrhosis of rats and guinea pigs. *Am. J. Path.*, 23: 159, 1947.
97. Calabresi, P. and Abelmann, W. H. Portocaval and portopulmonary anastomoses in Laënnec's cirrhosis and in heart failure. *J. Clin. Investigation*, 36: 1257, 1957.
98. Martin, W. J., Spittel, J. A., Morlock, C. G. and Baggenstoss, A. H. Severe liver disease complicated by bacteremia due to gram-negative bacilli. *Arch. Int. Med.*, 98: 8, 1956.
99. Ungar, H. Transformation of the hepatic vasculature of rats following protracted experimental poisoning with carbon tetrachloride. *Am. J. Path.*, 27: 871, 1951.
100. Lieber, M. M. The rare occurrence of metastatic carcinoma in the cirrhotic liver. *Am. J. M. Sc.*, 233: 145, 1957.
101. Bradley, S. E., Ingelfinger, F. J. and Bradley, G. P. Hepatic circulation in cirrhosis of the liver. *Circulation*, 5: 419, 1952.
102. Myers, J. D. The hepatic blood flow in Laënnec's cirrhosis with an estimate of the relative contributions from portal vein and hepatic artery. *J. Clin. Investigation*, 29: 836, 1950.
103. Vetter, H., Falkner, R. and Neumayr, A. The disappearance rate of colloidal radiogold from the circulation and its application to the estimation of liver blood flow in normal and cirrhotic subjects. *J. Clin. Investigation*, 33: 1594, 1954.
104. Krook, H. Circulatory studies in liver cirrhosis. *Acta med. Scandinav*, (supp.) p. 318, 1956.
105. Kessler, B. J., Leibler, J. B., Bronfin, G. J. and Sass, M. Hepatic blood flow and splanchnic oxygen consumption in alcoholic fatty liver. *J. Clin. Investigation*, 33: 1338, 1954.
106. Popper, H. and Schaffner, F. Liver: Structure and Function. New York, 1957. McGraw-Hill Book Co.
107. Calvy, G. L., Grant, R. N., Gliedman, M. L. and Leevy, C. M. Scientific exhibit. A. M. A. 106th annual meeting, New York, 1957.
108. Ebeling, W. C., Bunker, J. P., Ellis, D. S., French, A. B., Linton, R. R. and Jones, C. M. Management of patients with portal hypertension undergoing venous-shunt surgery. *New England J. Med.*, 254: 141, 1956.
109. Valdoni, P., Messini, M., Ricci, G. and Mancuso, M. La resezione epatica quale impulso rigenerativo nella terapia della cirroso. Prime osservazioni cliniche. *Epatologia*, 3: 1, 1957.
110. Schalm, L., Bax, H. R. and Mansens, B. J. Atrophy of the liver after occlusion of the bile ducts or portal vein and compensatory hypertrophy of the unoccluded portion and its clinical importance. *Gastroenterology*, 31: 131, 1956.
111. Madden, J. L., Lore, J. M., Jr., Gerold, F. P. and Ravid, J. M. Pathogenesis of ascites and considerations of its treatment. *Surg., Gynec. & Obst.*, 99: 385, 782, 1954.
112. Volwiler, W. In: Liver Disease. A Ciba Foundation Symposium. London, 1951. J. & A. Churchill, Ltd.
113. Baggenstoss, A. H. and Cain, J. C. The hepatic

hilar lymphatics of man: their relation to ascites. *New England J. Med.*, 256: 531, 1957.

114. Farber, S. J., Alexander, J. D. and Eichna, L. W. Renal hemodynamics and salt and water excretion during induced congestion of the inferior vena cava of man. *J. Clin. Investigation*, 30: 638, 1951.
115. Mangalik, V. S., Mehrotra, R. M. L. and Nayak, N. C. The pathogenesis of ascites in carbon tetrachloride cirrhosis. *J. Path. Bact.*, 73: 239, 1957.
116. Levitt, M. F., Turner, L. B. and Sweet, A. I. Effect of experimental venous obstruction on salt and water distribution and excretion in man. *J. Clin. Investigation*, 31: 885, 1952.
117. Dyrenfurth, I., Stacey, C. H., Beck, J. C. and Venning, E. H. Aldosterone excretion in patients with cirrhosis of the liver. *Metabolism*, 6: 544, 1957.
118. Iversen, P. The pathogenesis of ascites in liver disease. In: Liver Disease. A Ciba Foundation Symposium. London, 1951. J. & A. Churchill, Ltd.
119. Ratnoff, O. D. and Patek, A. J., Jr. Postnecrotic cirrhosis of the liver. *J. Chronic Dis.*, 1: 266, 1955.
120. Baggenstoss, A. H. Pathologic aspects of the late stages of viral hepatitis. In: Hepatitis Frontiers. Boston, 1956. Little, Brown & Co.
121. Transactions of the Fifth Meeting of the International Society of Geographical Pathology, Washington, D. C., 1954. *Schweiz. Ztschr. Path.*, 18: 382, 1955.
122. Hartroft, W. S. Experimental Hepatic Injury in Diseases of the Liver. Edited by Schiff, L. Philadelphia, 1956. J. B. Lippincott Co.
123. Gyorgy, P. and Goldblatt, H. Treatment in experimental dietary cirrhosis of the liver in rats. *J. Exper. Med.*, 90: 73, 1949.
124. Hartroft, W. S. The sequence of pathologic events in the development of experimental fatty liver and cirrhosis. *Ann. New York Acad. Sc.*, 57: 633, 1954.
125. Salmon, W. D. and Copeland, D. H. Liver carcinoma and related lesions in chronic choline deficiency. *Ann. New York Acad. Sc.*, 57: 664, 1954.
126. Schwarz, K. Factors protecting against dietary necrotic liver degeneration. *Ann. New York Acad. Sc.*, 57: 878, 1954.
127. Daft, F. S., Sebrell, W. H. and Lillie, R. D. Prevention by cystine or methionine of hemorrhage and necrosis in the liver of rats. *Proc. Soc. Exper. Biol. & Med.*, 50: 1, 1942.
128. Charnick, S. S., Moe, J. G., Rodnan, G. P. and Schwarz, K. A metabolic lesion in dietary necrotic liver degeneration. *J. Biol. Chem.*, 217: 829, 1955.
129. Schwarz, K. and Foltz, C. M. Selenium as an integral part of factor 3 against dietary necrotic liver degeneration. *J. Am. Chem. Soc.*, 79: 3293, 1957.
130. Abell, M. R. and Beveridge, J. M. R. Studies on hepatic necrosis induced by dietary means; conditions affecting production and prevention of massive liver necrosis. *Arch. Path.*, 52: 428, 1951.
131. Popper, H. and Schaffner, F. Nutritional hepatic injury. *Arch. Int. Med.*, 94: 785, 1954.
132. Popper, H., Szanto, P. B. and Elias, H. Transition of fatty liver into cirrhosis. *Gastroenterology*, 28: 183, 1955.
133. Seife, M., Kessler, B. J. and Lisa, J. R. Clinical, functional and needle biopsy study of liver in alcoholism. *Arch. Int. Med.*, 86: 658, 1950.
134. Davis, W. D., Jr. and Culpepper, W. S. Cirrhosis of the liver associated with alcoholism; report of acute exacerbation with serial liver biopsies. *Ann. Int. Med.*, 29: 942, 1948.
135. Schiff, L. Needle biopsy of the liver. Clinical applications in liver injury. Transactions of the Ninth Conference. New York, 1950. Josiah Macy, Jr. Foundation.
136. Popper, H., Szanto, P. B. and Parthasarathy, M. Florid cirrhosis. *Am. J. Clin. Path.*, 25: 889, 1955.
137. Albot, G., Herman, J., Faye, C. M. and Chardard, S. Pathogenesis of alcoholic hepatitis. *Am. J. Gastroenterol.*, 6: 53, 1957.
138. Albot, G., Schlumberger, C. S., Faye, C. M., Ruffino, J. and Raimbault, S. Le gros foie des alcooliques. Sa formule biologique et ses deux aspects histologiques. *Arch. mal. app. digest.*, 45: 585, 1956.
139. Best, C. H., Lucas, D. D. and Rideout, J. H. The lipotropic factors. *Ann. New York Acad. Sc.*, 57: 646, 1954.
140. Best, C. H., Hartroft, W. S., Lucas, C. C. and Rideout, J. H. Liver damage produced by feeding alcohol or sugar and its prevention by choline. *Brit. M. J.*, 2: 1001, 1949.
141. Klatskin, G., Gervin, H. M. and Krehl, W. A. Effects of prolonged alcohol ingestion on the liver of the rat under conditions of controlled adequate dietary intake. *Yale J. Biol. Med.*, 23: 317, 1951.
142. Vallee, B. L., Wacker, W. E. C., Bartholomay, A. F. and Hoch, F. L. Zinc metabolism in hepatic dysfunction. II. Correlation of metabolic patterns with biochemical findings. *New England J. Med.*, 257: 1055, 1957.
143. Davies, N. N. P. The pathology of dietary liver disease in tropical Africa. *Ann. New York Acad. Sc.*, 57: 714, 1954.
144. Scrimshaw, N. S., Behar, M., Arroyave, G., Tejada, C. and Viteri, F. Kwashiorkor in children and its response to protein therapy. *J. A. M. A.*, 164: 555, 1957.
145. Sepulveda, B., Hernandez de la Portilla, R., Rojas, E. and de J. Macias, J. Malnutrition and liver disease in Mexico. *Gastroenterology*, 33: 249, 1957.
146. Higginson, J., Grobbelaar, B. G. and Walker, A. R. P. Hepatic fibrosis and cirrhosis in man in relation to malnutrition. *Am. J. Path.*, 33: 29, 1957.
147. Payet, M., Peine, P. and Camain, R. Place de stéatose dans les cirrhoses dites nutritionelles des Africains adultes de Dakar. *Bull. et mém. de l'école preparatoire de méd. et de pharm. de Dakar*, 1: 29, 1952–1953.
148. Sharp, R. L., Snape, W. J. and Gilbert, P. D. Fatty metamorphosis of the liver associated with pancreatic calcification. *Gastroenterology*, 20: 647, 1952.
149. Hoffbauer, F. W., McCartney, J. S., Dennis, C.

and KARLSON, K. The relationship of chronic ulcerative colitis and cirrhosis. *Ann. Int. Med.*, 39: 267, 1953.

150. KLECKNER, M. S., JR. The liver in regional enteritis. *Gastroenterology*, 31: 416, 1956.
151. GALL, E. A. and LANDING, B. H. Hepatic cirrhosis and hereditary disorders of metabolism. *Am. J. Clin. Path.*, 26: 1399, 1956.
152. ZELMAN, S. Liver in obesity. *Arch. Int. Med.*, 90: 141, 1952.
153. WATSON, C. J., SUTHERLAND, D. and HAWKINSON, V. Studies of coproporphyrin; isomer distribution and per diem excretion of urinary coproporphyrin in cases of cirrhosis of liver. *J. Lab. & Clin. Med.*, 37: 8, 1957.
154. CAMERON, G. R. and KARUNARATNE, W. A. E. Carbon tetrachloride cirrhosis in relation to liver regeneration. *J. Path. Bact.*, 42: 1, 1936.
155. ERSHOFF, B. H. Conditioning factors in nutritional disease. *Physiol. Rev.*, 28: 107, 1948.
156. KLEMPERER, P. Die Leberveränderungen bei Schwammvergiftung. *Virchow's Arch. f. path. Anat.*, 227: 400, 1922.
157. KLECKNER, M. S., JR., BAGGENSTOSS, A. H. and WEIR, J. F. Hemochromatosis and transfusional hemosiderosis. *Am. J. Med.*, 16: 382, 1954.
158. RATHER, L. J. Hemochromatosis and hemosiderosis. *Am. J. Med.*, 21: 857, 1956.
159. FINCH, S. C. and FINCH, C. A. Idiopathic hemochromatosis, an iron storage disease. A. Iron metabolism in hemochromatosis. *Medicine*, 34: 381, 1955.
160. KARK, R. M. Two cases of aplastic anemia: one with secondary hemochromatosis following 290 transfusions in nine years, the other with secondary carcinoma of the stomach. *Guy's Hosp. Rep.*, 87: 343, 1937.
161. BLOCK, M., BETHARD, W. and JACOBSON, L. Secondary hemochromatosis. *J. Lab. & Clin. Med.*, 40: 781, 1952.
162. SCHWARTZ, S. O. and BLUMENTHAL, S. A. Exogenous hemochromatosis resulting from blood transfusions. *Blood*, 3: 617, 1948.
163. WYATT, J. P., MIGHTON, H. K. and MORAGUES, V. Transfusional siderosis. *Am. J. Path.*, 26: 883, 1950.
164. KENT, G., SCHRIFTER, H. and POPPER, H. Secondary hemochromatosis in blood dyscrasias with special emphasis on erythremic myelosis. *Am. J. Path.*, 32: 637, 1956.
165. CAPPELL, D. F., HUTCHISON, H. E. and JOWETT, M. Transfusional siderosis: the effects of excessive iron deposits on the tissues. *J. Path. Bact.*, 74: 245, 1957.
166. RICHTER, G. W. A study of hemosiderosis with the aid of electron microscopy. With observations on the relationship between hemosiderin and ferritin. *J. Exper. Med.*, 106: 203, 1957.
167. Observation of Kent, G.
168. UZMAN, L. The intrahepatic distribution of copper in relation to the pathogenesis of hepatolenticular degeneration. *Arch. Path.*, 64: 464, 1957.
169. BEARN, A. G. and KUNKEL, H. G. Abnormalities of copper metabolism in Wilson's disease and their relationship to the aminoaciduria. *J. Clin. Investigation*, 33: 400, 1954.
170. SCHEINBERG, I. H. and GITLIN, D. Deficiency of ceruloplasmin in patients with hepatolenticular degeneration (Wilson's disease). *Science*, 116: 484, 1952.
171. AHRENS, E. H., JR., PAYNE, M. A., KUNKEL, H. G., EISENMENGER, W. J. and BLONDHEIM, S. H. Primary biliary cirrhosis. *Medicine*, 29: 299, 1950.
172. GUTMAN, A. B. Drug reactions characterized by cholestasis associated with intrahepatic biliary tract obstruction. *Am. J. Med.*, 23: 841, 1957.
173. SONG, Y. S. Hepatic lesions in sickle cell anemia. *Am. J. Path.*, 33: 331, 1957.
174. EHRLICH, J. C. and RATNER, I. M. Congenital cirrhosis of the liver with kernicterus. *Am. J. Path.*, 31: 1013, 1955.
175. MCCULLOUGH, N. B. and EISELE, C. W. Brucella hepatitis leading to cirrhosis of the liver. *Arch. Int. Med.*, 88: 793, 1951.
176. KALK, H. Cirrhose und Narbenleber, 2nd ed. Stuttgart, 1957. Ferdinand Enke.
177. RATNOFF, O. D. Posthepatitic cirrhosis. *J. A. M. A.*, 165: 1098, 1957.
178. MONTENEGRO, M. R., DASILVA, L. C. and PONTES, J. F. An evaluation of the problems of hepatic cirrhosis as seen in São Paulo, Brazil. *Gastroenterology*, 33: 178, 1957.
179. ZIEVE, L., HILL, E., NESBITT, S. and ZIEVE, B. Incidence of residuals of viral hepatitis. *Gastroenterology*, 25: 495, 1953.
180. REAL-ENCINAS, R. Cirrhosis of the liver; incidence of various types at necropsy. Graduate School, University of Minnesota, Thesis, 1955.
181. STEIGMANN, F. Personal communication.
182. KUNKEL, H. G. and BEARN, A. G. Cirrhosis in young females: its possible relation to infectious hepatitis. In: Hepatitis Frontiers. Boston, 1956. Little, Brown & Co.
183. CRAIG, J. M., GELLIS, S. S. and HSIA, D. Y. Cirrhosis of the liver in infants and children. *Am. J. Dis. Child.*, 90: 299, 1955.
184. HEROLD, A. Contributions to the etiology, pathological anatomy and pathogenesis of liver cirrhosis in childhood. *Helvet paediat. acta*, 10: 427, 1955.
185. RUGGIERI, B. A., BAGGENSTOSS, A. H. and LOGAN, G. B. Juvenile cirrhosis. A clinicopathologic study of 27 cases. *J. Dis. Child.*, 94: 64, 1957.
186. LEEVY, C. M., GNASSI, A. M. and POLLINI, M. W. Clinical observations on hepatic fibrosis. *Arch. Int. Med.*, 96: 507, 1955.
187. SEIFE, M., KESSLER, B. J., HOFFMAN, J. and LISA, J. R. Clinical, functional and needle biopsy study of liver in pulmonary tuberculosis. *Am. Rev. Tuberc.*, 63: 202, 1951.
188. BOGLIOLO, L. The anatomical picture of the liver in hepatosplenic Schistosomiasis Mansoni. *Ann. Trop. Med.*, 51: 1, 1957.
189. RAGHEB, M. Schistosomiasis of the liver. Clinical, pathologic and laboratory studies in Egyptian cases. *Gastroenterology*, 30: 631, 1956.
190. MONTGOMERY, B. K. and ASZKANAZY, C. L. Postnecrotic cirrhosis in fibrocystic disease of pancreas. *Am. J. Clin. Path.*, 26: 630, 1956.
191. DE SANT'AGNESE, P. A. and BLANC, W. A. A distinctive type of biliary cirrhosis of the liver

associated with cystic fibrosis of the pancreas. *Pediatrics*, 18: 387, 1956.

192. Craig, J. M., Haddad, H. and Shwachman, H. The pathological changes in the liver in cystic fibrosis of the pancreas. *J. Dis. Child.*, 93: 357, 1957.
193. Stuart, K. L. and Bras, G. Veno-occlusive disease of the liver. *Quart. J. Med.*, 26: 291, 1957.
194. Bras, G., Berry, D. M. and Gyorgy, P. Plants as etiological factor in veno-occlusive disease of the liver. *Lancet*, 1: 960, 1957.
195. Selzer, G. and Parker, R. G. F. Senecio poisoning exhibiting as Chiari's syndrome. *Am. J. Path.*, 27: 888, 1951.
196. Benz, E. J. and Baggenstoss, A. H. Focal cirrhosis of the liver: its relation to the so-called hamartoma (adenoma, benign hepatoma). *Cancer*, 6: 743, 1953.
197. Parker, R. G. F. Fibrosis of the liver as a congenital anomaly. *Path. Bact.*, 71: 2359, 1956.
198. Krainer, L. Intrahepatic bile duct malformation associated with portal hypertension. *Gastroenterology*, 33: 265, 1957.
199. Martini, G. A. Cirrhosis of the liver in Osler's disease (telangiectatic cirrhosis). *Gastroenterologia*, 83: 157, 1955.
200. White, L. P., Phear, E. A., Summerskill, W. H. J. and Sherlock, S. Ammonium tolerance in liver disease: observations based on catheterization of the hepatic veins. *J. Clin. Investigation*, 34: 158, 1955.
201. Davidson, C. S., Summerskill, W. H. J. and Wolfe, S. J. Management of coma. In: Hepatitis Frontiers. p. 533. Boston, 1956. Little, Brown & Co.
202. Leevy, C. Portal hypertension. To be published.
203. Palmer, E. D. On the natural history of esophageal varices which are secondary to portal cirrhosis. *Ann. Int. Med.*, 47: 18, 1957.
204. Martini, G. A. Über Gefassveränderungen der Haut bei Leberkranken. *Ztschr. klin. Med.*, 153: 470, 1955.
205. Wallach, J. B., Hyman, W. and Angrist, A. A. The cause of death in patients with Laënnec's cirrhosis. *Am. J. M. Sc.*, 234: 56, 1957.
206. Chase, G., Martel, C. and Olivetti, R. Analysis of causes of death in Laennec's cirrhosis. *Am. J. M. Sc.*, 233: 259, 1957.
207. Allen, F. A., Carr, M. H. and Klotz, A. P. Decreased red blood cell—survival time in patients with portal cirrhosis. *J. A. M. A.*, 164: 955, 1957.
208. Ulevitch, H., Gall, E. A., Hoxworth, P. I., Schiff, L. and Graller, D. K. The importance of serial determinations of serum alkaline phosphatase in incomplete biliary tract obstruction. *J. Lab. & Clin. Med.*, 38: 693, 1951.
209. Watson, C. J. and Hoffbauer, F. W. The problem of prolonged hepatitis with particular reference to the cholangiolitic type and to the development of cholangiolitic cirrhosis of the liver. *Ann. Int. Med.*, 25: 195, 1946.
210. Schaffner, F. and Popper, H. Unpublished observations.
211. Gall, E. A. and Braunstein, H. Hepatitis with manifestations simulating bile duct obstruction. *Am. J. Clin. Path.*, 25: 1113, 1955.
212. Phillips, G. B. and Davidson, C. S. Liver disease of the chronic alcoholic simulating extrahepatic biliary obstruction. *Gastroenterology*, 33: 236, 1957.
213. Caroli, J. Les Lctères Par Retention. Paris, 1956. Masson et Cie.
214. Albot, G. La cholestase intrahepatique chronique. *Arch. mal. app. digest*, 46: 177, 1957.
215. Ricketts, W. E. and Kirsner, J. B. "Latent" portal cirrhosis. *Gastroenterology*, 17: 184, 1951.

Clinico-pathologic Conference

Asthma, Heart Murmurs, Cardiac Failure and Grand-Mal Seizures

STENOGRAPHIC reports, edited by Lillian Recant, M.D., and W. Stanley Hartroft, M.D., of weekly clinico-pathologic conferences held in the Barnes and Wohl Hospitals are published in each issue of the Journal. These conferences are participated in jointly by members of the Departments of Internal Medicine and Pathology of the Washington University School of Medicine and by Junior and Senior Medical students.

THIS white male engineer, sixty years of age, was admitted to Barnes Hospital for the fifth time on April 11, 1957. He died on April 20, 1957. His chief complaints were asthma and heart disease.

The patient had enjoyed good health in his youth, but believed that he was exceedingly nervous and once stated that he had had St. Vitus dance. At age twenty-three, he had inhaled a large concentration of phosphorus oxychloride, with resultant cough and hemoptysis for one week. During the subsequent five years he had worked in a cement plant during which time he was continually troubled with "colds." At age twenty-eight, while working as a field supervisor clearing the marshes in eastern Florida, his first episode of asthma developed which then troubled him from this time to his death.

At age forty-five (1941) a fistula developed in the right upper incisor which drained pus for five to six months. A heart murmur was heard for the first time by his physician, who stated that this murmur had not been present two years previously. At the time of the dental infection, an eighteen pound weight loss was noted and there was a decrease in exercise tolerance. The incisor was extracted after one year.

The patient was first admitted to Barnes Hospital in May, 1943, at age forty-seven, complaining of hazy vision in the right eye. Two weeks prior to admission he had awakened with marked blurring of vision in the right eye, unassociated with pain or fever. An ophthalmologist examined the eye and stated that he had suffered an occlusion of a retinal vessel.

Review of systems revealed the following: As a child the patient had had pneumonia. During the three years preceding this hospital admission there had been increased difficulty with asthma. He had never regained the eighteen pounds of weight lost two years previously. In 1923 an urethral stricture developed following the use of a mercurial ointment. Venereal disease was denied. In 1937, because of fever, he was treated for malaria with a single injection. In 1938, two-thirds of the left hand was amputated by machinery, necessitating skin grafting.

The family history was notable in that one sister was diabetic and that his parents lived past eighty years of age.

Physical examination revealed the temperature to be 37°c; pulse, 85; blood pressure, 122/70 mm. Hg. He was a moderately well developed, well nourished white man who did not appear ill; two-thirds of the left hand was amputated. The skin was clear. The lymph nodes were not enlarged. The pupils reacted to light and accommodation. Funduscopic examination of the right eye showed a pale, grayish discoloration of the upper two-thirds of the retina. The optic disc appeared normal. The arteries and veins were normal. There were no hemorrhages or exudates. The tonsils were enlarged but not inflamed and the pharynx was clear. The chest expanded normally with respiration. An occasional wheeze was heard in both lung fields. The heart was not enlarged to percussion. There was a normal sinus rhythm. A loud, diastolic, apical murmur was described by one observer. No abdominal organs or masses were palpable. Neurological examination was normal.

The laboratory data revealed the following: red blood cell count, 4.5 million/cu. mm.; hemoglobin, 14 gm./100 ml.; white blood cell count, 5,000/cu. mm. with basophils, 2 per cent; eosinophils, 3 per cent; metamyelocytes, 3 per

cent; polymorphonuclear leukocytes, 63 per cent; lymphocytes, 20 per cent; and monocytes, 9 per cent. The urinalysis showed a specific gravity of 1.016; reaction, pH 4.5; protein and glucose, negative. A few urate crystals were seen. The stool was guaiac negative. The Kahn test was negative. Non-protein nitrogen was 19 mg./100 ml.; fasting blood sugar, 80 mg./100 ml. Electrocardiogram showed a PR interval of 0.21-seconds with "probable myocardial damage on basis of low T-1." Gastrointestinal series and intravenous cholecystogram were normal. Basal metabolic rate was +8 per cent. Blood cultures showed no growth on two occasions. Skin tests showed slight erythema to house dust.

While in the hospital for ten days, eye complaints diminished and other medical complaints were investigated. The patient underwent urethral dilatation. He was discharged to undergo desensitization to house dust. The discharge diagnosis was probable arterial retinal occlusion.

The patient was admitted to the hospital for the second time from December 29, 1949 to January 21, 1950. During the six year interval, he had continued to have occasional nocturnal paroxysms of wheezing and cough, which had increased in severity in the two years preceding this admission. Episodes of wheezing, dyspnea and cough lasted from three to four weeks with the production of mucopurulent sputum following "colds." Such an episode had begun one month prior to this admission and had not responded to the administration of antihistiminics, epinephrine by nebulizer, aminophyllin intravenously, or to three injections of penicillin. Edema of the ankles or dyspnea did not occur between the attacks of wheezing.

Physical examination disclosed a temperature of 37.7°c.; pulse, 110; respirations, 28; blood pressure, 140/90 mm. Hg. The patient appeared dyspneic and orthopneic from moderately severe asthma. There was decreased vision in the right eye with the right optic disc pale. Slight erythema of the throat was present. There was a notable increase in the A-P diameter of the chest. The chest was hyperresonant and the diaphragms were low. There was marked wheezing and a prolonged expiratory phase. The heart was not enlarged to percussion and the rhythm was regular. The heart sounds were of good quality. A grade 2 systolic blowing murmur was heard at the apex by two observers. No diastolic murmur was noted. The liver was palpable 2 cm. below the right costal margin.

The laboratory studies revealed a hemoglobin of 16.5 gm./100 ml., red blood cell count of 5.3 million/cu. mm. and a white blood cell count of 13,400/cu. mm. with 4 per cent eosinophils and an otherwise normal differential. A trace of protein was found in the urine along with 10 to 15 white blood cells per high power field in the centrifuged sediment. Smear of the sputum showed many neisserian organisms, a few gram-positive cocci in chains, and a few gram-negative rods. There were 83 per cent polymorphonuclear leukocytes and 17 per cent eosinophils in the sputum. Sputum culture revealed a pure growth of neisserian organisms. Throat culture grew a few colonies of alpha hemolytic streptococci. Skin tests revealed slight sensitivity to varied antigens. Roentgenograms of the chest demonstrated generalized pulmonary emphysema of minimal degree and bilateral apical pleural thickening. Films of the paranasal sinuses revealed thickening of the mucous membranes of both maxillary antra with a fluid level on the left. Ethmoid and sphenoid sinusitis were noted. An electrocardiogram was indeterminate with a PR interval of 0.16 seconds.

On bed rest, an elimination diet, administration of aminophyllin intravenously and penicillin intramuscularly, sedation, iodides and antihistaminics, the patient rapidly improved, with clearing of the chest and decrease in fever. Asthmatic symptoms recurred with cessation of treatment, necessitating reinstitution of therapy with aminophyllin. The patient had a low-grade fever during his course in the hospital but was discharged improved after twenty-three days.

The third admission to the Barnes Hospital was from February 13 to February 21, 1957. In the intervening seven years the patient had continued to suffer recurrent episodes of cough and wheezing, especially at night, following upper respiratory infections and physical exertion. Cortisone had been administered intermittently for the preceding six years. Eighteen months prior to this admission the patient had been treated steadily with prednisone. Symptoms of congestive heart failure, notably increased dyspnea at rest and upon exertion, had commenced approximately six months prior to this admission, progressing to orthopnea, paroxysmal nocturnal dyspnea and edema of the ankles during the last two weeks. Chest roentgenograms

had demonstrated an increase in heart size, both right and left ventricular enlargement, and were suggestive also of left auricular enlargement. Pulmonary congestion and bilateral pleural effusion were observed on films taken eight days before admission and the patient was admitted for therapy.

Vital signs were as follows: temperature, 37°C.; pulse, 90; respirations, 20; blood pressure, 120/80 mm. Hg. The patient was alert, cooperative and denied any distress. However, he utilized the accessory muscles of respiration vigorously. The optic fields and fundi were as previously noted. The lymph nodes were not enlarged. Examination of the chest revealed an increased A-P diameter. Wheezes were heard throughout the lung fields. There were no signs of fluid. The left cardiac border was percussed 9 cm. to the left of the mid-sternal line in the fifth intercostal space. The rhythm was regular. A harsh, grade 4 systolic murmur was heard loudest at the apex but also at the aortic area with radiation to the carotid arteries, and along the left cardiac border from the pulmonary area to the apex. A2 was diminished but louder than P2; M2 was loud. A low pitched pre-systolic murmur was heard at the apex by several observers, but denied by others. There was no thrill and no venous distension. The soft, non-tender edge of his liver was felt 5 cm. below the right costal margin. The spleen tip was palpable 2 cm. below the left costal margin. There was no cyanosis, clubbing or edema. Rectal examination was normal.

The laboratory findings showed a normal hemogram. Urinalysis was normal with the exception of 5 to 10 white blood cells and an occasional red blood cell per high power field. Non-protein nitrogen was 34 mg./100 ml. Cholesterol was 256 mg./100 ml. Serum CO_2, chloride, albumin, globulin, acid and alkaline phosphatase were all normal. Numerous eosinophils and polymorphonuclear leukocytes were found in his sputum. A few beta hemolytic streptococci and a moderate growth of alpha hemolytic streptococci and paracolon bacilli were cultured from the sputum. An occasional coliform bacillus and a heavy growth of paracolon bacilli were cultured from the urine. The venous pressure was 55 mm. of saline and the circulation time was prolonged to thirty seconds (arm to tongue). The electrocardiogram revealed an abnormal form of ventricular complex suggestive of myocardial damage with the P-R interval 0.20 seconds. (The T wave was inverted in standard lead 1, and in leads V1 through V5.) Roentgenograms of the chest revealed right and left ventricular and left atrial enlargement, pulmonary vascular congestion with an interval decrease from the previous examination, an interval decrease in the left pleural effusion and resolution of the right pleural effusion.

The patient was allowed a 3 gm. salt diet and unrestricted activity. He was maintained on prednisone, 10 mg. daily, and digitalized with the leaf preparation. Concomitant with the loss of several pounds in weight there was marked subjective improvement insofar as respiratory symptoms were concerned. The circulation time declined to seventeen seconds. He was discharged on this regimen, taking 0.1 gm. of digitalis daily. It was planned that steroids would be withdrawn gradually. The many cardiologists who saw the patient concurred in deferring cardiac catheterization and surgery because of the patient's age and the associated pulmonary disease.

The fourth admission to the Barnes Hospital was from March 10 to March 24, 1957. The patient had continued to do well. Prednisone had been decreased to 5 mg. daily. At 4 A.M. on the morning of admission he was awakened from his sleep by a grand mal seizure which lasted two minutes, without incontinence. Following this episode the patient could not be aroused for fifteen minutes. Four similar episodes occurred in the subsequent seven hours. During one such episode the patient's wife noted the heart beating slowly and faintly. She also recalled a similar convulsion in August, 1956.

On physical examination the temperature was 38.7°C.; pulse, 115; respirations, 12 to 16; and blood pressure, 166/92 mm. Hg. The patient was drowsy but could recall all events up to the twenty-four hours preceding the seizures. The lungs were clear. Initially the cardiac rhythm was irregular, but within fifteen minutes it was regular. The previously described systolic murmurs were noted. No diastolic murmurs were heard. The liver was palpable 5 cm. below the right costal margin. The spleen was not felt. The right pupil was larger than the left. There was generalized hyperreflexia of the left arm and leg. A positive Babinski sign was present on the right side and questionably on the left. The neck was supple.

Laboratory data were as follows: a hemogram was normal; proteinuria was 1 plus; the non-

protein nitrogen was 37 mg./100 ml. The serum sodium, potassium, CO_2, chloride and proteins were within normal limits. Serum calcium was 9.5 mg./100 ml. and phosphorus was 4.5 mg./100 ml. Cephalin cholesterol flocculation test was 2 plus, the thymol turbidity test was 8.3. The transaminase was 77 units. Examination of the urine for phenylpyruvic acid was negative. The Schwartz-Watson test was negative. An electrocardiogram revealed auriculoventricular nodal rhythm with AV dissociation and periods of sinus rhythm with incomplete AV block and Wenckebach AV conduction; left ventricular enlargement, digitalis effect and frequent auricular premature contractions with a rate of 120. Chest roentgenograms revealed cardiomegaly with an interval decrease in size. A phonocardiogram showed a systolic murmur in all ausculatory areas, most intense in the aortic area. Because of this localization and its diamond shaped configuration, aortic stenosis was thought to be its origin.

The patient was thought to have suffered Stokes-Adams syncope, secondary to AV dissociation and digitalis intoxication. Digitalis was discontinued. Isuprel® (isoproterenol HCl) was administered by linguets every three hours, along with barbiturates and potassium chloride administered orally. Within eighteen hours, sinus tachycardia was present with frequent auricular premature contractions. At forty-eight hours the electrocardiogram returned to its pre-convulsive appearance.

X-ray films of the skull were normal. A lumbar puncture revealed an initial pressure of 148 with final pressure of 92 mm. of water. The protein was 49 mg./100 ml.; the sugar 78 mg./100 ml.; and the chlorides, 124 mEq./L. The Wassermann and colloidal gold tests were normal. Transaminase on the cerebral spinal fluid was 5 units. An electroencephalogram showed intermediate slow dysrhythmia, without localization. Hypoxia, rather than a primary convulsive disorder, was thought to be the most likely basis of the electroencephalographic changes. It was concluded clinically that the patient had a vascular basis for the hemiparesis and convulsion. He was given dilantin® (diphenylhydantoin sodium) and anticoagulated with dicumarol. Administration of digoxin, 0.25 mg. daily, was reinstituted prior to discharge and prednisone therapy was maintained.

The final admission to Barnes Hospital was from April 11 to April 20, 1957. On the regimen mentioned the patient had done moderately well at home. He began to complain of sweating and occasional mild chills with questionable nightly fevers. Several days prior to admission he was told that he had a "strep" throat for which he was given penicillin intramuscularly and chlortetracycline orally. For a few nights there had been increased dyspnea despite the use of an oxygen tent. Dyspnea increased on the day of admission.

Vital signs were as follows: temperature, 37.6°C.; pulse, 92 and regular; respiration, 26 and labored; and blood pressure, 160/70 mm. Hg. The patient appeared acutely and chronically ill. He was sweating profusely. Coarse rhonchi and wet rales were heard throughout both lung fields with both inspiratory and expiratory wheezes. The cardiac rhythm was regular with occasional ventricular premature contractions. The liver was again palpable 3 cm. below the right costal margin.

The laboratory data showed a normal hemogram with 6 per cent eosinophils. The non-protein nitrogen was 22 mg./100 ml. Blood cultures on two occasions failed to reveal growth. Roentgenograms of the chest revealed marked pulmonary congestion, particularly in the upper lung fields. The electrocardiogram was essentially unchanged.

The patient was febrile with the oral temperature ranging about 38°C. with an occasional spike to 39°C. Some objective improvement was noted in the lung fields. Six days following admission in the early morning he was found breathing rapidly and shallowly; he appeared cyanotic, responding only with effort. He was wheezing audibly. The blood pressure was 130/70 mm. Hg and the pulse 110. Aminophyllin given intravenously had little effect and within fifteen minutes the patient was noted to have markedly constrictive and non-reactive pupils. He did not respond to stimuli and appeared flaccid. Electrocardiographic changes were ascribed to the tachycardia or possibly to acute cor pulmonale due to pulmonary embolism or acute asthma. The serum potassium was 5.8 mEq./L. When the oxygen being administered was decreased to 2 L./minute the patient became more responsive but remained semi-stuporous for one to two hours. Within eight hours he was lucid and comfortable. All medications except dicumarol, dilantin® and prednisone were discontinued. The next morning the non-protein nitrogen was 35 mg./100 ml.; the CO_2 was 33.6; the sodium,

135; potassium, 4.7; and chloride, 92 mEq./L. The episode was variously ascribed to hypoxia, aortic stenosis, hyperkalemia and cerebral vascular disease. Fever persisted and three days later the patient again became unresponsive with rapid, labored, shallow respirations. The lung fields were tight with much wheezing. He became extremely toxic with the temperature rising to 41°C., not declining with sponging and intravenously administered fluid. The heart sounds became barely audible and following a few agonal gasps he died at 11:30 A.M.

CLINICAL DISCUSSION

DR. EDWARD REINHARD: This patient died at the age of sixty. His first admission to Barnes Hospital was in 1943 when he was forty-seven years old. He gave a history of pneumonia and an indefinite history of "St. Vitus dance," both in childhood. He had an acute exposure to phosphorus oxychloride at age twenty-three which caused a profound illness of brief duration. We do not know whether or not he had any chronic exposure to this compound, but he did work in a cement plant for a period of five years, from age twenty-three to twenty-eight, and we are told that he had constant colds during this time. He had a urethral stricture at age twenty-seven, which was said to be non-venereal in origin. Asthma, first occurring at age twenty-eight, continued to be a prominent feature of his medical history until death. He had one episode of fever, which he was told might be malaria, at age forty. His hand was amputated as the result of an industrial accident at age forty-one. At age forty-five, he had an infected tooth with purulent drainage from a fistula for one year, at the end of which time the tooth was finally extracted. During this time, heart murmurs were first described. We are told by the private physician that he definitely did not have any cardiac murmurs prior to this time.

The patient was first admitted to the hospital with an arterial occlusion in his eye, thought to be caused by an embolus. Starting in 1943 occasional episodes of nocturnal paroxysmal wheezing and coughing developed which progressively increased in severity. He was ultimately treated with intermittent courses of cortisone which seemed to control the symptoms. He received continuous prednisone therapy from August, 1955 to February, 1957. He had symptoms of recurrent congestive heart failure from August, 1956 until February, 1957 when he was again admitted to the hospital. He had a good response to cardiotherapy, at least during the initial episode. However, he had severe congestive heart failure during the terminal part of his illness. He had a convulsion in August, 1956 seen only by his wife. It was apparently very mild but he later had severe seizures. During the last three weeks of his life he had sweats, chills and fever. Three days prior to his death, he had shallow rapid respirations and cyanosis. He became unresponsive and his temperature rose to 41°C. The changes in physical findings were striking over the seventeen year period. There was a progressive increase in heart size. He had wheezing throughout this time with an increased diameter of the chest suggestive of some degree of pulmonary emphysema. He had signs of heart failure at different times. He had fever, cyanosis, an episode of unresponsiveness and great respiratory distress terminally.

On his first admission to Barnes Hospital, only one examination of the heart is recorded which states that the patient had a loud apical diastolic murmur. On all subsequent admissions, a systolic murmur of varying intensity was heard at the apex and at the base but a diastolic murmur was heard only occasionally and it was never audible to all observers. The systolic murmur was considered to be compatible with aortic stenosis. Dr. Bukantz, Dr. Bercu and Dr. Binder described a definite diastolic murmur at the apex which they considered to be mitral stenosis. Dr. Bukantz thought there might also be the murmur of mitral insufficiency. Dr. Massie examined this patient repeatedly and did not hear the diastolic murmur. Either he had a very indistinct diastolic murmur heard only by some people or the murmur was intermittent. Now, Dr. Humphrey, would you review the roentgenograms?

DR. HARVEY A. HUMPHREY: The first examination in 1950 showed that the heart was not grossly enlarged. There was a prominent left auricle so that I think the x-ray evidence favors rheumatic heart disease. By August, 1955 the size of the heart had definitely increased. There was also some increase in density suggesting calcification in the mitral ring or valve plus evidence of right ventricular enlargement and left auricular enlargement. The changes together with the mitral calcification, are interpreted as evidence of mitral stenosis. The esophagus was displaced. There was questionable calcification in the region of the aortic

valve. In May, 1956, the patient was in heart failure, as manifested by pulmonary vascular congestion, increase in heart size, and some evidence now of left ventricular enlargement. In February, 1957, there was a sizeable pleural effusion on the left, and pulmonary congestion with an interval increase in the size of the pulmonary vessels attributable to pulmonary hypertension. There was also fracture of the left seventh rib attributed to trauma. On the final admission, the heart had decreased in size. The lungs had cleared. Some calcification was evident in the aortic arch. In summary, there was progressive cardiomegaly with evidence of left auricular, and right and left ventricular enlargement. These findings can be interpreted on the basis of mitral stenosis and aortic disease, with pulmonary congestion and edema.

DR. REINHARD: Dr. Price, would you discuss both the electrocardiograms and the phonocardiograms?

DR. KENNETH C. PRICE: The electrocardiogram of April 17, 1957 is quite representative of the patient's tracings taken in the last few months of life. There are symmetrical T-wave inversions in $V_1V_2V_3$ which suggest the possibility of myocardial ischemia or right ventricular strain. The ST-T changes in $V_4V_5V_6$ and the increased voltage of the R-waves are characteristic of left ventricular enlargement and digitalis effect. The P-waves in II, III and AVF are relatively high as compared to the QRS voltage. (These are often referred to as "P-pulmonale" waves.) The P-waves are notched. The atrioventricular conduction is prolonged. In summary, the significant electrocardiographic findings suggest: (1) left ventricular enlargement and digitalis effect, (2) myocardial ischemia or right ventricular strain, and (3) possible cor pulmonale.

The phonocardiogram records a rhomboid systolic murmur occurring in the first half of systole in the aortic area. In the mitral area a murmur is also recorded in the first half of systole. The murmur is loud and the frequency is variable. As heard by the ear, it should have "musical characteristics. The phonocardiogram therefore, suggests aortic stenosis of unknown degree and mitral insufficiency. There is no diastolic murmur recorded.

DR. REINHARD: The one phonocardiograph recording does not, of course, eliminate the possibility of an intermittent diastolic murmur. Dr. Goldman, would you comment on the history of exposures to phosphorus oxychloride and to dust in the cement plant? Could this have contributed to his asthmatic difficulties?

DR. ALFRED GOLDMAN: Phosphorus oxychloride, $POCl_3$, to which he was exposed, is a colorless fuming liquid with a pungent odor and it is quite toxic and corrosive. It is soluble in water at which time it decomposes and heat is evolved. We do not know the exact amount of exposure our patient had. Certainly it would be possible that he had irritation in his pharynx and his larynx, perhaps some edema, and a bronchitis and a bronchial pneumonia might have developed at that particular time. I was unable to find any cases of lung lesions due to phosphorus oxychloride in the literature. However, there is a very similar compound used for the same purpose, a chlorinating agent; it is phosphorus chloride, PCl_3, and is definitely hazardous. As a result of acute exposure, conjunctivitis, pharyngitis and bronchitis may occur and, following repeated exposures over a period of one to two years, asthma and emphysema may result. The answer to how deleterious the $POCl_3$ fumes might have been, would be dependent on more intimate knowledge of the amount of exposure, and the symptoms following the injury. As to the cement hazard, cement workers do not get pneumoconiosis. Gardner [*1*] in 1939 reported on the study of some 2,200 men in the Portland cement industry. Persons who were exposed for a period of five years showed nothing in the lungs. Those who were exposed for many years showed linear reticulated shadows and occasionally nodulation. There is silicon dioxide in the cement, but it does not exist in the free state, so one does not get silicosis. As to the incidence of colds and coughs in the cement workers, I would say there probably is an increase over that occurring in workers of the same age group in other industries.

DR. REINHARD: Dr. Bukantz, is it possible that exposure of the bronchi to caustic chemicals could lead later on in life, to the intrinsic type of asthma?

DR. SAMUEL C. BUKANTZ: My answer would be yes, but I do not think I could support it with any valid data. In many people who have had exposures to such irritant substances as the World War I poison gases intrinsic asthma has developed many years later, as it did in this patient. Incidentally, I would like to correct

[1] GARDNER, L. U. Survey of 2,200 cement workers. *J. Indust. Hyg. & Toxicol.*, 21: 7, 1939.

the record in one respect; this patient was symptom free for some twelve to fifteen years following exposure to the $POCl_3$. I think the same sort of process may affect the irritated bronchus and lung that affects the pericardium and myocardium in valvotomized patients in whom the postvalvotomy syndrome develops. This may be an autosensitization.

DR. REINHARD: Do you think a chronic infectious bronchitis develops in the damaged bronchi and leads to asthma?

DR. BUKANTZ: Yes, infection may play a part.

DR. REINHARD: It seems clear that this patient had asthma for some years before any manifestations of congestive heart failure developed. He certainly did not have cardiac asthma alone. He may, however, have had some aggravation of his asthma by decompensation later. Dr. Bukantz, is it not true that congestive failure is apt to cause an aggravation of the manifestations of asthma?

DR. BUKANTZ: I suppose that this would be true particularly in the person in whom left ventricular failure develops. I do not think it is true of cor pulmonale when an increase of the dyspnea does not at first seem to occur.

DR. REINHARD: Dr. Massie, do you think this patient had rheumatic heart disease?

DR. EDWARD MASSIE: Yes. He gave a history of St. Vitus dance in childhood and we are told that a murmur was heard at the age of forty-five. The heart murmurs certainly could be compatible with rheumatic disease. We all agreed that the patient had a loud systolic murmur at the aortic area and at the apex. These sounded like the murmurs of aortic stenosis and mitral insufficiency. The only difference of opinion was whether or not the murmur of mitral stenosis was also present. Mitral insufficiency alone could have accounted for the left auricular enlargement noted on roentgenogram.

DR. REINHARD: Dr. Smith, if we assume for the sake of argument that the patient did have a diastolic murmur at the apex in addition to the systolic murmur described, would it necessarily indicate the presence of a mitral lesion or might it be secondary to the supposed aortic lesions?

DR. JOHN R. SMITH: Diastolic murmurs due to aortic regurgitation may be evanescent. The murmurs may be present one day and absent on the next. Apparently low grade changes in the dynamics in aortic insufficiency cause the murmurs to come and go. It is quite possible that an aortic lesion was present. However, mitral valve disease, with predominant regurgitation, seems probable.

DR. REINHARD: You would agree with Dr. Massie about the type of heart disease?

DR. SMITH: Yes, rheumatic heart disease is the best working diagnosis.

DR. REINHARD: When a patient has valvular heart disease with no classical history for rheumatic fever, we always have to consider other etiologies. Do you think, Dr. Bing, that fibroelastosis should be considered here?

DR. RICHARD BING: Since fibroelastosis may be manifested by heart murmurs and markedly enlarged hearts in adults, it should be considered. There are two forms of fibroelastosis, one that makes its appearance in infancy and usually results in a rapid death of the patient, and the other which appears in adult life and can lead to a condition similar to that encountered in this patient.

DR. REINHARD: In a study of twenty patients with endocardial fibroelastosis, Thomas [2] found that nine had definite cardiac murmurs and eleven had no murmurs. Of these murmurs, most of them were systolic and only one patient had a short diastolic rumble. Dr. Bing, do you believe this is a very likely diagnosis?

DR. BING: The most likely diagnosis would be rheumatic heart disease, but fibroelastosis is a very distinct possibility. One should also consider other more rare conditions such as tumor of the heart and amyloidosis of the heart. These can produce heart murmurs. The mechanism is that very often amyloidosis leads to changes in the endocardium with shrinkage, and then to constriction of the valve rings.

DR. REINHARD: Dr. Smith, would you attribute the progressive cardiomegaly to the valvular heart disease or do you think there was superimposed cor pulmonale?

DR. SMITH: It would be difficult to separate them because there were evidences of both lesions. Asthma and chronic pulmonary disease may have accounted for some of the difficulty, added to which there is the probability of mitral valve disease.

DR. REINHARD: Dr. Humphrey and I reviewed the roentgenograms together and we did not think there was good radiological evidence of marked pulmonary emphysema. Now, let us consider the so-called grand-mal seizures and the subsequent terminal episode of

[2] THOMAS, W. Endocardial fibroelastosis. *New England J. Med.*, 251: 327, 1954.

unresponsiveness and semi-stupor. Dr. Smith, some of these episodes were attributed to Stokes-Adams syncopy.

DR. SMITH: Stokes-Adams seizures may occur under circumstances involving marked slowing of ventricular rate (and consequent curtailment of minute cardiac output), often by the abrupt appearance of AV block, or by falling idioventricular rate in an established AV block. In this case the attacks were attributed by some observers to digitalis intoxication, and consequent AV dissociation. Syncopal seizures of different mechanism may occur in patients with aortic stenosis. I have suspected (without final proof) that in some patients disastrous syncopal episodes may be invoked by ordinary parasympathetic stimuli, especially in the presence of anoxia, when the effects of parasympathetic stimulation can become greatly magnified, whether upon the heart, the cerebral circulation, or the peripheral vasculature.

DR. REINHARD: Dr. Bukantz, you took care of this patient during several of his hospital admissions. Were these five episodes in a period of seven hours true convulsive seizures, or were they really syncopal attacks?

DR. BUKANTZ: No one witnessed them but the wife. She did not phone for any medical assistance until the sixth, or the final episode had occurred. When he was seen, he was so obviously in a postictal state of depression, that I would think it almost certain that he had had grand mal seizures. There had been no tongue biting; and one of the striking things about our personal experience with this patient was the difficulty of ascertaining when left ventricular failure took over and when bronchial asthma quit, or when they were combined. The most striking period of benefit that he exhibited was at a time when he was receiving both digitalis and steroids. He was asymptomatic and had clear lung fields. We then slowly withdrew the steroids and the episode of the grand-mal seizures occurred about ten days later. I had not seen him in the interim and the seizures developed at a time when he again was wheezing a great deal. It was evident that he was suffering both from left ventricular failure and recurrent bronchial asthma. We all believed that the return of the bronchial asthma and the advent of digitalis toxicity with changing ventricular rhythms accounted for the anoxia which precipitated the convulsive state.

DR. REINHARD: The patient had high fever only during the last three or four weeks of his life. Although he had previous episodes of fever, it is at least possible that most of these might have been associated with upper respiratory infections and were not directly related to the terminal febrile illness. Assuming this to be so, let us consider briefly what the terminal episode of fever might have been due to, noting particularly the causes of fever in association with valvular heart disease. Any patient with valvular heart disease can get any kind of fever. But there are certain diseases to which they are peculiarly susceptible. Active rheumatic fever did not seem very likely. Bacterial endocarditis was considered a distinct possibility. Three blood cultures during his terminal eleven days in the hospital were all negative. Drug fever is another possibility, but he was not taking any drugs likely to cause fever. Pulmonary embolism should also be mentioned. Dr. Smith, could the roentgenographic appearance of the chest with congestion in the upper part of the lung be a manifestation of pulmonary emboli, or do you agree with Dr. Humphrey that pulmonary congestion was more likely?

DR. SMITH: One should think of pulmonary embolism. The event can occur suddenly, and does not necessarily appear on the x-ray film. All the symptoms displayed by this man could have been excited by embolism, although this possibility remains uncertain.

DR. REINHARD: Congestive heart failure can cause a low grade fever, but not a temperature of 41°C. unless there were complications. Hemoglobin pneumonia sometimes causes fever in patients with mitral disease. That apparently was not the case here. The patient had no hemoptysis. Disseminated lupus and other collagen diseases were considered by several people seeing this patient to be the possible underlying cause for this clinical syndrome. Dr. Moore, do you want to defend any type of collagen disease as contributing to this patient's illness, particularly the terminal febrile illness?

DR. CARL V. MOORE: I would not like to defend any of the collagen diseases as likely possibilities.

DR. REINHARD: Atrial myxomas do cause fever. Ball-valve thrombi probably could not have produced an illness lasting as long as this patient's over-all illness, but could have occurred as a terminal event. The long duration of this illness does not in any way exclude atrial myxoma as a possible cause for his long standing cardiac murmurs. Patients have survived forty-

three years with myxoma of the atrium, at least they have had a syndrome lasting a long time which could have been due to an atrial myxoma and they did have myxoma at autopsy. Many cases have been reported in which the clinical syndrome, presumably attributable to the myxoma lasted over a period of three, four or even five years. My final diagnoses are bronchial asthma; pulmonary emphysema without cor pulmonale; a cardiac lesion, probably rheumatic valvulitis. However, myxoma of the auricle, a very rare lesion could explain the findings. A patient with myxoma was described in whom one of the outstanding features was convulsive epileptiform seizures. He had fever, murmurs and everything that this patient had. The diagnosis is attractive, but statistically, I have to put it second. Fibroelastosis is unlikely. The patient had congestive heart failure contributing to the syndrome, probably with a superimposed severe lung infection although the x-ray findings could be attributed only to congestion in the lungs. I believe the seizures were due to cerebral anoxia, secondary to the cardiac lesion. Dr. Hartroft will present and discuss both the gross and microscopic findings.

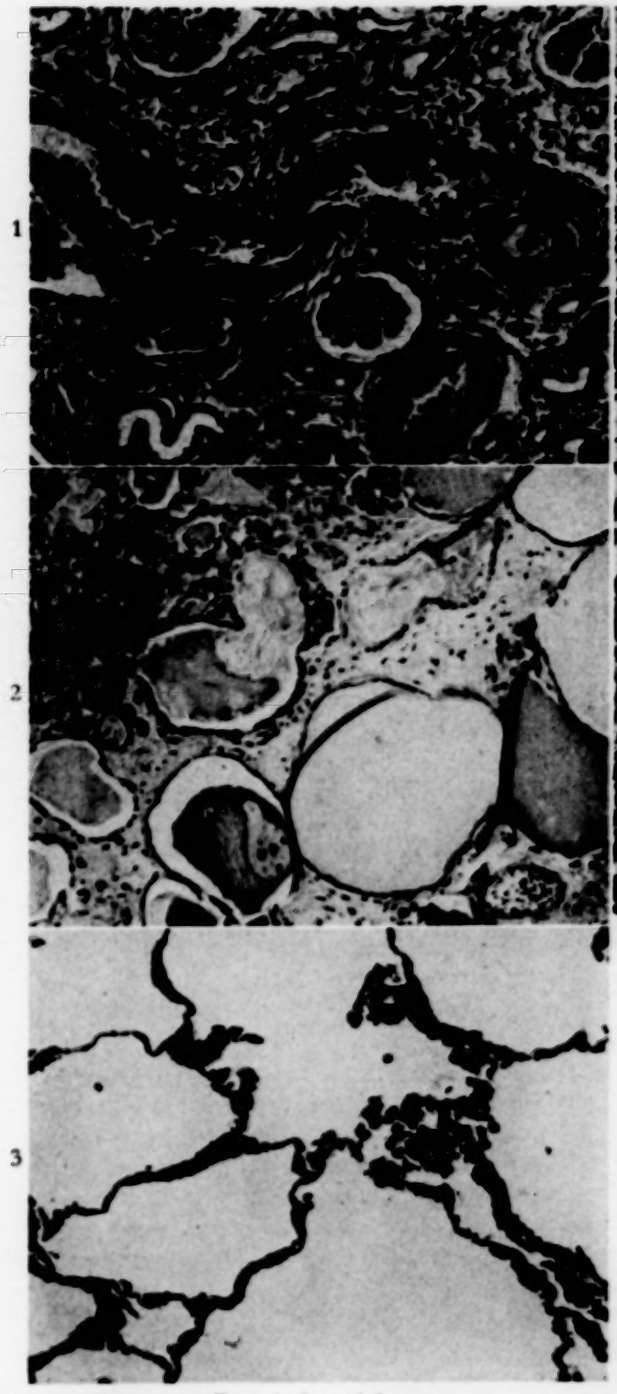
PATHOLOGIC DISCUSSION

DR. W. STANLEY HARTROFT: This sixty year old, one-handed man exhibited a great variety of pathologic lesions which were present in the gastrointestinal tract, the urogenital system, the endocrines and in the liver, spleen, lungs and heart. I will deal with them in that order. The brain showed no abnormalities to which we could attribute functional significance; only those non-specific general changes to be expected in a patient with this history of general anoxia. A small diverticulum was present in the descending portion of the colon. Its neck opening into the lumen of the bowel was filled with inspissated secretion as was the entire lumen of the diverticulum. Its wall exhibited evidence of a low grade chronic inflammation in the wall. In the colon at the splenic flecture was a small polyp quite benign in nature. In the wall of the sigmoid and the upper part of the rectum were many polymorphonuclear leukocytes and he obviously had suffered from both acute and chronic prostatitis. Further up in the sigmoid was found another benign polyp. In the appendix, we observed a small growth at its tip. It proved to be a small carcinoid tumor.

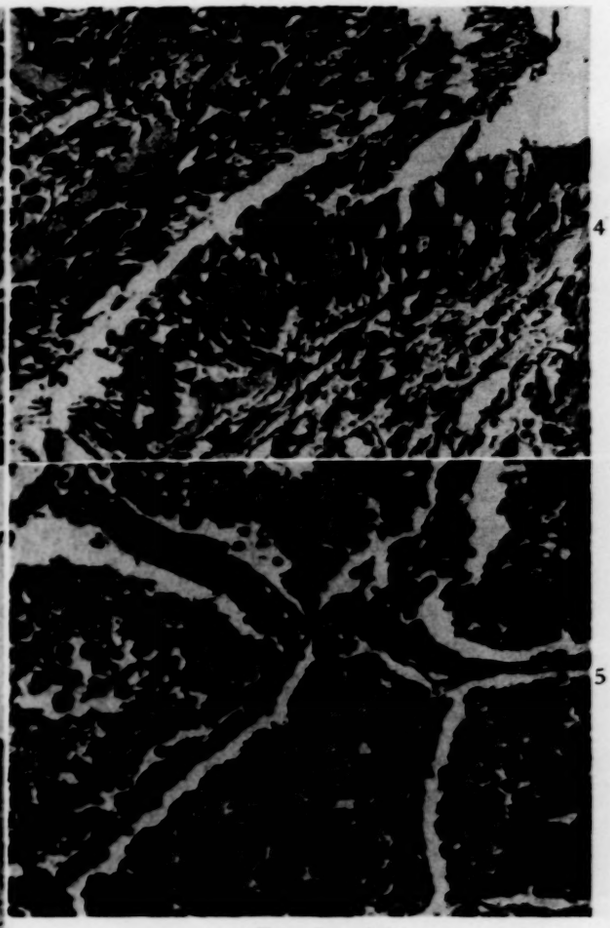
In the prostate was found a well differentiated carcinoma of the prostate which although small was well developed. The rest of the urogenital system was within normal limits but for a healed infarct of the kidney. (Fig. 1), with loss of tubules and very tortuous thickened vessels representing endarteritis.

In the adrenal artery, there was severe depletion of lipid in the zona fasciculata and zona glomerulosa. In the pituitary (Fig. 2) in the pars intermedia, colloid-filled cysts were present but they left plenty of functioning pituitary tissue in the anterior portions. In the pancreas, we found focal areas of acinar atrophy in which only coiled ducts persisted in a sea of collagen, the waves of which lapped on the shores of islets which persisted. The liver exhibited all the classic changes, grossly and microscopically, of congestive fibrosis and early cirrhosis.

In the lungs, we found alternating areas of collapse and focal type of emphysema. (Fig. 3.) Mucus was prominent in all sections of bronchi. Mucus was the feature that captured the attention and imagination of the prosector, Dr. W. Talbert at the time of the autopsy for it was so abundant and tenacious, filling every portion of the bronchial tree. The walls of the bronchi afforded examples of the classic lesions of asthma (Fig. 4) with hyperplasia of epithelium and metaplasia. The basement membrane was thickened. Beneath this thick hyaline basement membrane there was infiltration of eosinophils. In other regions of the lung were prominent thickened alveolar walls in which accumulations of red blood cells and hemosiderin laden macrophages suggested a circulatory disturbance. (Fig. 5.) Still other alveoli were filled with masses of fibrin in which were enmeshed leukocytes and red blood cells. The lesions were those of bronchopneumonia with organization and fibrosis. In the heart there were areas of fibrous scarring somewhat ovoid in shape. They might be the end result of rheumatic myocarditis. Grossly, the striking feature was the hypertrophy of the left ventricle with enlargement and dilation. The endocardium here was thickened by fibrous tissue. The right ventricle also was hypertrophied and dilated. The cusps of the aortic valve were calcified but not stenosed. The mitral valve was however, of greater interest. Rupture of the chordae tendineae of the posterior cusps of this valve had occurred. Their loose ends were thickened and fibrosed indicating rupture had taken place a long time before death. The papillary muscle attached to the anterior cusp



Figs. 1, 2, and 3.



Figs. 3 and 4.

was not ruptured and was hypertrophied, whereas the muscle of the ruptured chordae was atrophic and small. The rupture, we think, explains a good deal of the clinical signs and symptoms. Rupture of chordae tendineae is a distinct clinical and pathological entity [3–7].

It is seen not too uncommonly as a part of the picture of a terminal acute or subacute bacterial endocarditis when there is erosion of the valve. But there are a number of cases reported in the literature of rupture of chordae tendineae in the absence of acute or subacute bacterial endocarditis and in the absence of major lesions in the other parts of the valves. Two of the reported cases have been definitely described with good evidence of rheumatic endocarditis and four questionably. The mitral ring of our case was calcified as well as the posterior cusp. If rupture

[3] Corvisart, J. N. Essai sur les Maladies et les Lesions Organiques du Coeur et des Gross Vaisseaux, 3rd ed., pp. 420–421. Paris, 1818. Mequignon-Marvis.

[4] Dry, T. J. et al. Certain cardiac lesions which simulate mitral stenosis. *Heart Bull.*, 4: 70, 1955.

[5] Bailey, O. T. and Hickman, J. B. Rupture of mitral chordae tendineae. (Report of seven cases in which there was no bacterial endocarditis.) *Am. Heart J.*, 28: 578, 1944.

[6] Frew, H. W. O. Rupture of the chordae tendineae following scarlet fever. (A case in a child.) *Glasgow M. J.*, 115: 195, 1931.

[7] Frothingham, C. and Hase, G. M. Rupture of normal chordae tendineae of the mitral valve. *Am. Heart J.*, 9: 492, 1934.

of the chordae is complete as in this case, then its papillary muscle will become atrophic and the other papillary muscle will become hypertrophic. But if only a part of the attachment of the chordae tendineae of the valve which has been ruptured is torn, why both muscles will become hypertrophic. Incompetence of the valve, of course, produces cardiac hypertrophy, failure and dilation as we saw. Rupture of this type, as in our case, is commonest in middle aged elderly people. The predominant symptoms clinically are those of congestion and edema. Hypertension need not be present. Congestive cirrhosis is present in most reported cases because the vascular disturbance is an ideal setup for producing this lesion in the liver. Cardiac murmurs may be helpful diagnostically. The precordial systolic murmur should be loud and harsh with the maximum sound at the apex along the left sternal border. A thrill may be felt in the same area. Sometimes, an apical diastolic murmur also is present. A systolic pulsation of left atrium is considered very diagnostic when present. The murmurs are thought to develop rather suddenly. In our case the murmur did appear rather quickly fifteen years before death. The etiology and pathogenesis of these ruptures of chordae tendineae have been generally thought to be secondary to some other pathologic lesion because it does not seem reasonable that it should happen to a healthy valve. We might suspect rheumatic endocarditis of the mitral valve as predisposing here in combination with the dyspnea from asthma. Repeated respiratory infections for seven years explain his organizing bronchopneumonia and emphysema. I think this case is a good example of how disease in one organ will react on another organ and intensify symptoms.

Final Anatomical Diagnoses: Primary: Tenacious, glairy plugs in all branches of tracheobronchial tree (clinical diagnosis of intrinsic asthma, thirty years); emphysema, all lobes, slight; trabeculations of the tracheal mucosa; organizing bronchopneumonia, upper lobes, both lungs, advanced; follicular hyperplasia of the spleen; congestion of the spleen and kidneys and of the mucosa of the stomach, cecum and rectum; arteriolar nephrosclerosis, slight; hypertrophy of the heart, mainly left ventricular (465 gm.); ruptured, healed, chordae tendineae of the free edge of the posterior leaflet of the mitral valve (systolic murmur, clinical diagnosis of aortic stenosis); thickening of the remaining chordae tendineae to the posterior leaflet of the mitral valve; calcification near the base of the posterior leaflet of the mitral valve at the insertion of the remaining chordae tendineae; calcification of the mitral valve ring near the posterior leaflet of the mitral valve; dilatation and hypertrophy of the left atrium, moderate (mitral valve insufficiency); thickening of the endocardium of the left atrium, slight; focal fibrosis of the myocardium. Accessory: Arteriosclerosis of the abdominal aorta and coronary arteries, marked; of the descending thoracic aorta, moderate; of the cerebral, renal, splenic, pulmonary and aortic arch, slight; sclerosis of the bases of the cusps of the aortic valve, slight; adhesions of the commissures of the cusps of the aortic valve, slight; lipid infiltration of the anterior leaflet of the mitral valve, slight; focal fibrosis of the epicardium of the right ventricle, slight; calcified nodule, right lower lobe of the lung; apical pleural scar, bilateral; fibrous adhesions of the pleura of the left lower lobe of the lung, slight; infarct of the left kidney, small, healed; absence of the left hand (history of traumatic amputation, 16 years); healed scar left lower quadrant of the abdomen (full thickness of skin graft for the hand); carcinoid tumor of the appendix, small; atrophy of the liver, slight; thrombosed internal hemorrhoidal veins; prostatic calculi, one, small; occult carcinoma of the posterior lobe of the prostate, microscopic; pedunculated polyps of the colon, small (two); diverticulosis of the distal transverse descending and sigmoid colon, slight; osteoarthritis of the dorsal and lumbar spine, slight; osteoporosis, slight; lipid depletion of the adrenals, slight.

Case Reports

Multiple Aneurysmal Formation*

An Elastic Tissue Defect

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ANEURYSMAL formation of the aorta in all age groups has been well documented [*1,2*]. Comprehensive reports in the literature noted involvement of all segments of the aorta. In a few of these cases multiple aneurysms involving more than one segment were observed.

Aneurysmal formation of the coronary arteries alone is an even greater rarity. Since the first case report of Bougon in 1812 [*3*], forty-eight cases have been recorded. An excellent and comprehensive review was that of Scott [*4*] which confirmed the rare occurrence of this entity and the even greater rarity of such aneurysms accompanied by aneurysms of the aorta [*5,6*]. Most coronary artery aneurysms have been described as single, only nine being recorded as multiple. We have found no case report of coronary artery aneurysm accompanied by aneurysmal formation of any visceral vessel.

Recently, we had occasion to perform an autopsy on a forty-four year old white man who had multiple aneurysmal formations of the left and right coronary arteries, multiple aneurysms of the aorta in all its segments and in its major abdominal branches, and, in addition, an aneurysm of the hepatic artery in the liver parenchyma. Such multiple aneurysmal formation in one patient is unusual indeed.

FIG. 1. The heart, showing multiple aneurysms of the anterior descending and circumflex branches of the left coronary artery.

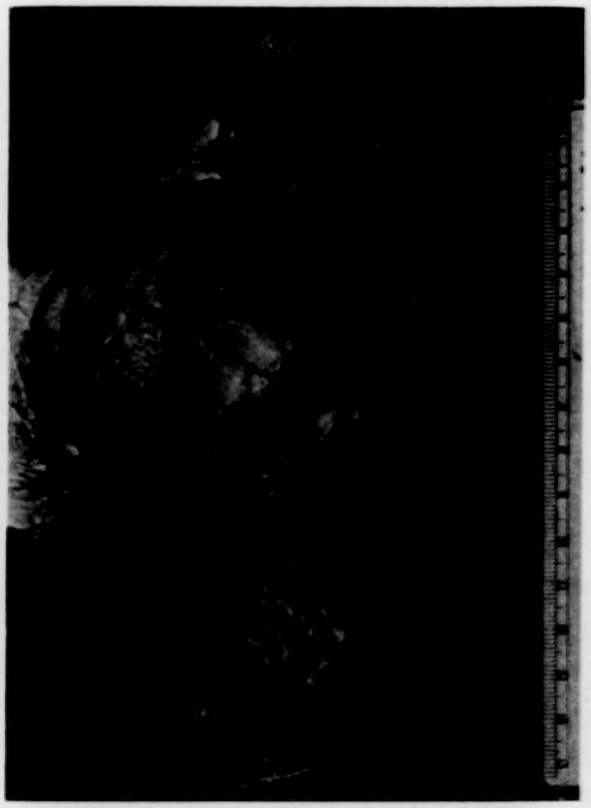

FIG. 2. Anterior descending branch of the left coronary artery, showing aneurysm and proximal fresh thrombus formation.

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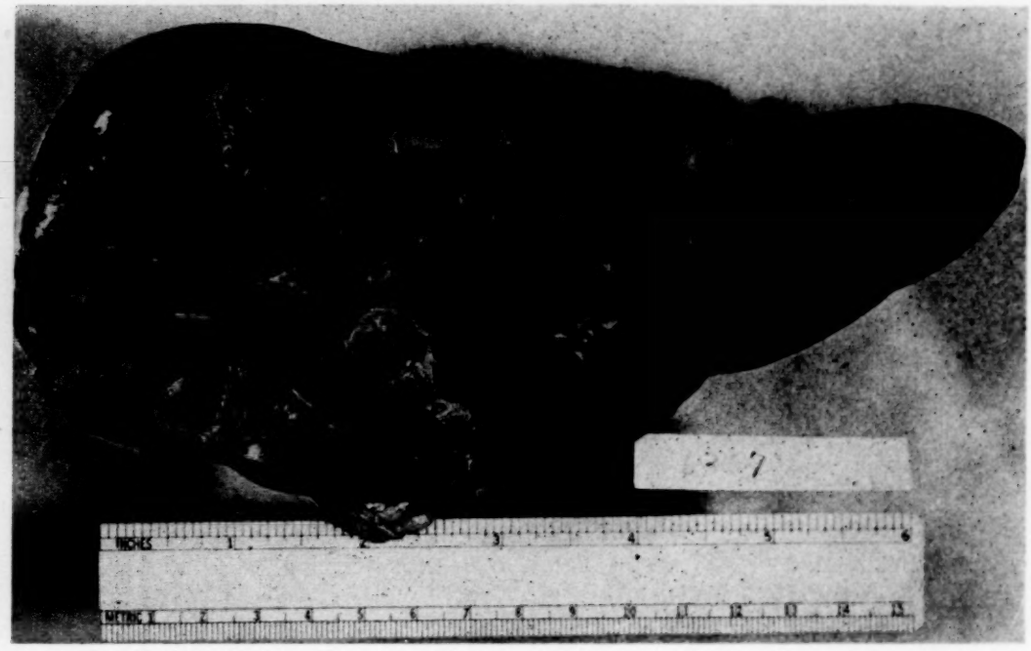

FIG. 3. Cut section of the liver, revealing aneurysmal formation of the hepatic artery.

CASE REPORT

A. A., a forty-four year old white man, native of Puerto Rico, was admitted to the Lincoln Hospital for the first time on March 3, 1956 with the following symptoms of one week's duration: shortness of breath on exertion, pain (non-radiating) over the right upper quadrant of the abdomen, and cough productive of blood-tinged sputum. The past history revealed three episodes of pneumonia in the last ten years, and ill-treatment as a prisoner of war in Japan from 1941 to 1945. There was a 10 pound weight loss in the last six months. The familial history was non-contributory. Since his release from military service in 1945 the patient had worked as an able-bodied seaman in the merchant marine.

On admission, physical examination revealed a well developed, but chronically ill, white man in acute distress with a temperature of 99°F.; blood pressure, 130/85 mm. Hg; and respirations, 24/minute. The pertinent cardiac findings were distention of jugular veins bilaterally and cardiac enlargement to the right and left by percussion. There was normal sinus rhythm. Examination of the lungs revealed impaired resonance and subcrepitant rales at both bases. The liver was palpated five fingerbreadths below the right costal margin. The impression on admission was congestive heart failure of unknown etiology —possibly beri-beri heart disease.

Examination of the urine revealed a 3-plus albuminuria with specific gravity of 1.015, and a sediment containing numerous polymorphonuclear leukocytes. The hemoglobin was 9.5 gm. per cent; red cell count, 4,240,000 per cu. mm.; white cell count, 8,600 per cu. mm. with a normal differential count. Liver profile studies revealed: serum alkaline phosphatase, 6.3 Bodansky units; serum albumin, 3.5 gm. per cent; globulin, 3.2 gm. per cent; cholesterol, 157 mg. per cent; cholesterol ester, 85 mg. per cent; direct bilirubin, 0.2 mg. per cent; indirect bilirubin, 0.4 mg. per cent; thymol turbidity, 7 units; serum urea nitrogen determination was 14 mg. per cent; blood sugar (fasting) was 105 mg. per cent. The Wassermann test was negative. The venous pressure was 240 mm. water, the decholin® circulation time was twenty-three seconds. The electrocardiogram was interpreted as consistent with Wolff-Parkinson-White syndrome.

The patient was digitalized and treated intensively for congestive heart failure. The temperature course was afebrile, blood pressure readings were recorded between 124 and 130 mm. Hg systolic and 85 to 90 mm. Hg diastolic. In spite of intensive treatment the patient died in congestive heart failure. The clinical impression was myocarditis of unknown etiology.

Postmortem examination was performed eight hours after death. The gross anatomic findings were: coronary thrombosis of the anterior descending branch of the left coronary artery; recent myocardial infarction; myofibrosis cordis; adherent mural thrombus at the apex of the left ventricle; multiple aneurysmal formations of the circumflex branch of the left coronary artery; aneurysmal formation of the anterior descending branch of the left coronary artery; pericarditis; aneurysm of the interventricular septum and apex of the left ventricle; saccular aneurysm of the transverse arch of the aorta; multiple saccular aneurysms of the abdominal aorta above the iliac branches; aneurysm of the hepatic artery in the parenchyma of the liver; multiple aneurysmal forma-

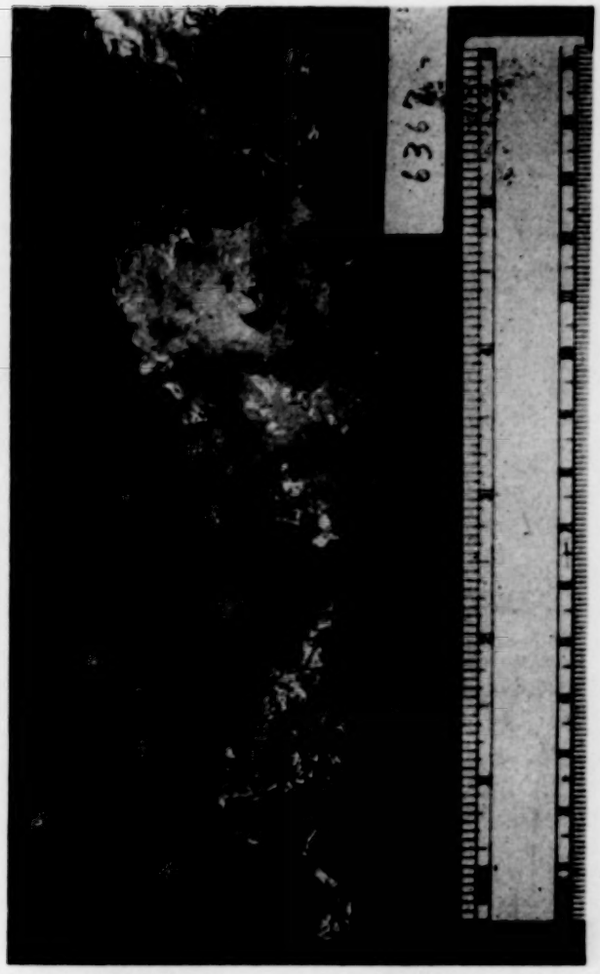

Fig. 4. Multiple aneurysmal formation in the major branches of the aorta. The celiac artery is at top center, immediately below is the superior mesenteric artery.

Fig. 5. The adventitial surface of the abdominal aorta, showing multiple aneurysmal formation in the major branches. The celiac artery is at top center, the superior mesenteric artery is just below it.

tions of the abdominal aorta at the branches of the celiac plexus, superior mesenteric artery, right renal artery, testicular artery; multiple old infarctions of both kidneys; advanced chronic passive congestion of liver; chronic passive congestion of both lungs; arteriosclerosis of the cerebral vessels at the base of the brain.

The heart was enlarged in its transverse diameter and weighed 525 gm. (Fig. 1.) There was increased serous fluid in the pericardial sac and fibrinous adhesions were found between the visceral and parietal layers of the pericardium at the apex. The cavity of the right ventricle was markedly reduced in size, to a crescent-shaped area, due to bulging of the interventricular septum and apex. The wall was thin and fibrotic in this latter area. Attached to this area was an adherent thrombus of red-grey color 3.2 by 2.6 cm. In the wall of the left ventricle anterior and laterally was a linear zone of fibrosis 1.5 by 0.6 cm. There was also noted in the myocardium at the apex and slightly posteriorly an area of yellow-white discoloration surrounded by a thin hyperemic zone.

On examination of the anterior descending branch of the left coronary artery an aneurysm was noted 1.4 cm. in diameter located 4 cm. from the left coronary orifice. Proximal to the aneurysm was a recent adherent thrombus of red-grey structure. (Fig. 2.) The aneurysm contained laminated yellow-grey material adherent to the vessel walls. In the circumflex branch of the left coronary artery, two aneurysmal formations of similar structure were noted, each 0.9 cm. in diameter, 3.2 cm. and 4 cm. from the left coronary orifice. In the right coronary artery, 1.6 cm. distal to the right coronary orifice, a 1 cm. aneurysm of laminated grey-white substance was noted adherent to the vessel walls.

Examination of the aorta revealed a saccular aneurysm of 3.5 cm. in length distal to the origin of the left common carotid artery. It contained grey-yellow gelatinous material adherent to the wall. Calcified plaques were noted in the wall.

The significant findings in the abdominal viscera included aneurysmal formation of the hepatic artery,

FIG. 6. The elastic tissue fibers of the medial coat of the thoracic descending aorta are widely separated from each other and fragmented. Verhoeff stain.

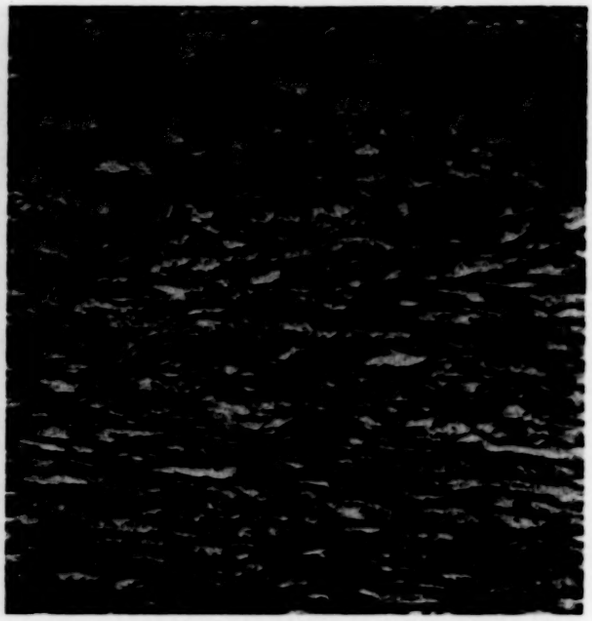

FIG. 7. The same area as Figure 6, showing zones of metachromasia corresponding to degeneration and fragmentation of elastic tissue fibers. Toluidine blue stain.

of 2.4 by 2.0 cm. in dimension, in a major branch. (Fig. 3.) Marked passive congestion also was present in this liver which weighed 1,750 gm. The kidneys weighed 225 gm. and 200 gm., respectively. They showed irregular depressed scars beneath the capsule, indicative of old infarction. In the abdominal aorta there were aneurysmal formations immediately distal to the origin of celiac artery, 1 cm. in diameter, with calcified, rigid walls. The superior mesenteric, right renal, and testicular arteries showed similar aneurysmal formations but with varying degrees of calcification and rigidity of the wall. (Figs. 4 and 5.) There was also a saccular aneurysm of the abdominal aorta distal to the inferior mesenteric artery, 2.2 cm. in length. Similar aneurysms (three in number) above the bifurcation were also noted. All these aneurysms contained calcified, rigid walls.

The constant histologic feature encountered in this case was fraying and fragmentation of the elastica, with collections of metachromatic substance. (Figs. 6 and 7.) This was noted in the aorta and in its major branches. Toluidine blue stain revealed these substances, but periodic acid-Schiff stain gave a negative reaction. Sections taken through the areas of aneurysmal formation revealed obvious sclerosis and hyalinization beneath the intima, with medial degeneration. Toluidine blue stain in these areas failed to reveal metachromatic substances. Verhoeff stain in these areas disclosed nests of fragmented and ruptured elastica in the areas adjacent to the aneurysm (Figs. 8 and 9); the walls of the latter showed no elastica.

In addition, sections of aorta also showed subintimal zones of lipid, which were not constantly present, however, at all levels. Sections through the areas of aneurysmal formation were likewise inconstant in showing this subintimal lipid. The coronary aneurysms and hepatic aneurysm revealed hyalinization and destruction of media. Hemorrhagic extravasation with blood pigment and chronic inflammatory cells were observed about the adventitia. Calcification of the walls was also noted.

The findings in the heart muscle indicated recent infarction and old myofibrosis cordis. Examination of the kidney revealed old infarcts due to embolization from the adherent mural thrombus of the left ventricle. Finally, there was passive congestion of the lungs and liver.

COMMENTS

Of paramount interest in this case is a consideration of the possible etiologic factor or factors responsible for such multiple aneurysmal formation in a forty year old man. In a review of 369 necropsies of aortic aneurysms during the period from 1892 to 1953, Brindley and Sternbridge [*1*] found that 10.6 per cent of their patients had multiple aneurysms involving more than one aortic segment. Syphilis was far less frequent in these multiple aneurysms than in

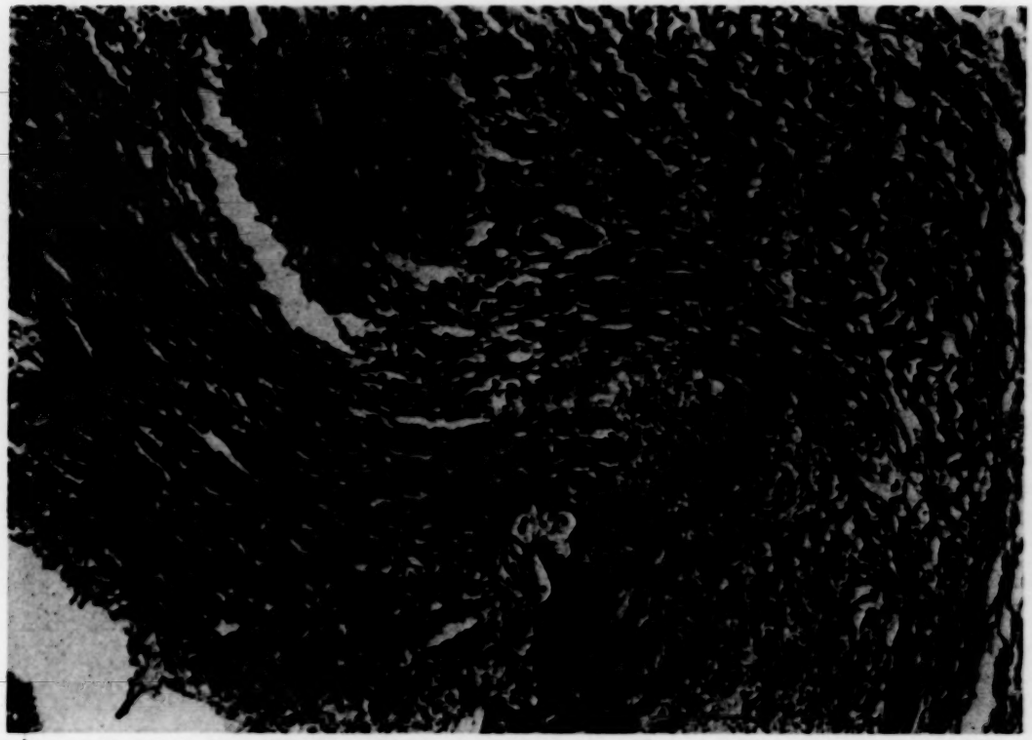

FIG. 8. Section of the abdominal aorta at the periphery of an aneurysm, showing fragmentation of elastic fibers and widening of the spaces between the fibers (left). There is disappearance of these fibers at the site of the aneurysm. Verhoeff stain.

FIG. 9. Section at junction of an aneurysm and the grossly normal wall of the renal artery at its origin from the abdominal aorta, showing thickening, degeneration and rupture of elastic fibrils, with pools of amorphous substance between ruptured fibers (lower left). Verhoeff stain.

single aneurysms. The important etiologic factors in these multiple aneurysms were arteriosclerosis and cystic medial necrosis. These authors indicate that during the last ten year period of their survey the incidence of cystic medial necrosis was 22 per cent as compared with the initial years of their survey when the same lesion was reported in only 1 per cent of the cases. These cases are exclusive of Marfan's syndrome in which the aortic lesion is that of cystic medial necrosis. Gore [*2*] reported sixteen cases of aortic aneurysm in the age group nineteen to thirty-five in which the lesion was medial degeneration involving the elastica. He further claimed that the lesion he noted coincides with the cystic medial necrosis observed by many in Marfan's syndrome [*7,8*]. Many authors [*9–11*] also described a cystic medial necrosis of the aorta in the younger age group, similar to the lesion described by Gore. Wolff [*12*] reported an unusual case of diffuse elastic tissue destruction with mucoid accumulations in the aortic media of a twelve day old infant. This lesion was associated with somatic defects including cardiac hypertrophy and widening of the aortic and pulmonic valve cusps. Similar lesions in association with coarctation of the aorta have been reported in young people [*13,14*].

It would seem, therefore, that this lesion is more prevalent than originally described by Erdheim [*15*]. It is not limited to the older age group but occurs frequently in young people. In the latter, there is rapid development of signs and symptoms in physically active young persons with a subsequent downhill course, as contrasted to the older age group in which the natural history of the disease is more protracted.

Despite the large number of case reports and the variety of methods used by various investigators in pursuing the study of this lesion, there is little agreement regarding its genesis and nature. Schlichter [*16*] was able to produce necrosis of the aortic media and rupture by sudden interference with adventitial vascularization. With slower interference he was able to show mucoid cystic changes in the media. He therefore concluded that interference with vascularization of the aorta determines the site and extent of the severity of the lesions. By means of diet and induction of renal insufficiency in experimental animals Holman [*17*] was able to demonstrate aneurysmal lesions of the aorta, pulmonary artery and coronary arteries. These lesions start as edema and swelling of the intercellular tissue of the large elastic arteries and advance to necrosis with cellular infiltration. In the advanced lesions there is fragmentation and lysis of the elastic framework. Similar lesions even more typical of cystic medial necrosis, showing elastolysis and fragmentation, were produced by feeding rats a diet containing 50 per cent Lathyrus odoratus seeds [*18,20*]. In these animals multiple aneurysmal lesions were produced in the aorta, pulmonary arteries and coronary arteries. Leary and Weiss [*19*] likewise demonstrated medionecrosis of the aorta in a rabbit fed a high cholesterol diet. This case was unique. However they were consistently able to produce medial necrosis in these animals, with a tendency to calcification, by giving them massive doses of vitamin D. Such lesions were unaccompanied by dissecting aneurysms.

The incidence of cystic necrosis and medial degeneration of the aorta has been reported to be increased in pregnancy. Schnitker and Bayer [*21*] reported forty-nine cases of dissecting aortic aneurysm with cystic medial degeneration. In twenty-four of these cases (49 per cent) there was an associated pregnancy. In these cases hypertension was not a factor, nor was labor; there were no congenital anomalies. The altered blood lipids coincident to pregnancy in a patient vulnerable to cystic medial necrosis has been thought to be the mechanism of dissection in such a high incidence of cases. Bauersfeld [*13*] also reported on the incidence of pregnancy and dissecting aneurysm of the aorta. He concluded similarly that syphilis, arteriosclerosis, hypertension and congenital defects, although they may be present in any given case, are not *per se* responsible for dissection. He postulated a hormonal change weakening the aortic wall.

Last in our consideration is the very comprehensive review of elastic tissue by Hass [*22*]. In a discussion of the effects of age on elastic tissue, he indicated that "a relentless course is followed, sometimes retarded and sometimes hastened, but always in an established sequence." The progressive change is a loss of elastic tissue in all vascular elastic networks, with retention of collagen. Further, with distention of elastic tissue and decrease in hydration, their colloidal dispersion is interfered with, resulting in masses of elastica prone to calcific and iron impregnation. This process is independent of atherosclerosis.

It is interesting to note that in 1889 Thoma [*23*] postulated a molecular disintegration of the

coats of the aorta which would predispose to aneurysm and was subsequently able to demonstrate microscopic degenerative changes in early cases of angiomalacia (1920). Costa [*24*] in a study of human embryos found the arterial walls rich in mucoid substance but this substance disappeared after the first month in the elastic arteries, remaining only in musculoelastic arteries. If it is supposed that the defect in the elastica, with fragmentation and pooling of metachromatic substances, is one of metabolism or nutrition, or both, the widespread extent of the aneurysms can be understood.

Cystic medial necrosis with fragmentation of elastica may be considered to be due either (1) to over-production of normally present mucoid substance, with resulting encroachment of elastic and muscle tissue, (2) to the result of a degenerative process which may be secondary to abnormal hormonal or metabolic processes, or (3) to a consequence of the progressive morphologic changes of aging.

A review of the literature on coronary artery aneurysms revealed that arteriosclerosis and congenital defects were considered to be the etiologic agents in a great number of cases. In regard to the former, the frequency of arteriosclerosis without aneurysmal formation is so great, that it is difficult to accept arteriosclerosis alone as an etiologic factor. In regard to the latter, Scott [*4*] suggested that "possibly it would be wiser to leave this group unclassified until further evidence can be obtained to indicate more definitely what the etiology is." It may indeed be wiser to leave these lesions unclassified.

These lesions of multiple aneurysms have a common denominator, namely destruction of elastic tissue, hyalinization and eventual calcification. If we consider that the average age of these patients (with coronary aneurysms) was only forty-seven, and if we do not accept arteriosclerosis as the etiology, we may speculate that these lesions are the result of an elastic tissue defect. Aneurysmal formation is the natural sequel.

SUMMARY

A case of multiple aneurysmal formation involving the aorta and its major branches, the coronary vessels and the hepatic artery, is presented. The possible etiologic factors are reviewed. The common feature of elastic tissue defect is suggested as the etiology.

REFERENCES

1. Brindley, P. and Sternbridge, V. A. Aneurysms of the aorta. *Am. J. Path.*, 32: 67, 1956.
2. Gore, I. Dissecting aneurysms of the aorta in persons under forty years of age. *Arch. Path.*, 55: 1, 1953.
3. Bougon. Cited by Scott, D. H. [*4*].
4. Scott, D. H. Aneurysm of the coronary arteries. *Am. Heart J.*, 36: 403, 1948.
5. Cox, R. L. and Christie, C. D. Aneurysm of the coronary arteries. Report of a case. *Am. J. M. Sc.*, 180: 37, 1930.
6. Manohar, K. D. An aneurysm of a coronary artery. *Arch. Path.*, 26: 1131, 1938.
7. MacLeod, M. and Williams, A. W. The cardiovascular lesions in Marfan's syndrome. *Arch. Path.*, 61: 143, 1956.
8. Tobin, J. R., Jr., Bay, E. B. and Humphreys, E. M. Marfan's syndrome in the adult. *Arch. Int. Med.*, 80: 475, 1947.
9. Magner, D. and McKenzie, H. F. Cystic medionecrosis of the aorta. *Arch. Path.*, 44: 485, 1947.
10. Rottino, A. Medial degeneration of the aorta as seen in twelve cases of dissecting aneurysm. *Arch. Path.*, 28: 1, 1939.
11. Sailer, S. Dissecting aneurysms of the aorta. *Arch. Path.*, 33: 704, 1942.
12. Wolff, K. Cited by Sailer, S. [*11*].
13. Bauersfeld, S. R. Dissecting aneurysm of the aorta; a presentation of fifteen cases and a review of the literature. *Ann. Int. Med.*, 26: 873, 1947.
14. Harrison, F. F. Coarctation of the aorta of the adult type associated with cystic degeneration of the media in the first portion of the arch. *Arch. Path.*, 27: 742, 1939.
15. Erdheim. Cited by Sailer, S. [*11*].
16. Schlichter, J. G. Experimental medionecrosis of the aorta. *Arch. Path.*, 42: 182, 1946.
17. Holman, R. L. Necrotizing arteritis in dogs related to diet and renal insufficiency. *Am. J. Path.*, 19: 977, 1943.
18. Ponseti, I. V. and Baird, W. A. Scoliosis and dissecting aneurysm of the aorta in rats fed with Lathyrus odoratus seeds. *Am. J. Path.*, 28: 1059, 1952.
19. Leary, T. and Weiss, S. Dissecting aneurysm of the aorta in experimental atherosclerosis. *Arch. Path.*, 29: 665, 1940.
20. Walker, D. G. and Wirtschafter, Z. T. Histopathogenesis of aortic aneurysms in the lathyrus fed rat. *Arch. Path.*, 61: 125, 1956.
21. Schnitker, M. A. and Bayer, C. A. Dissecting aneurysm of the aorta in young individuals, particularly in association with pregnancy. *Ann. Int. Med.*, 20: 486, 1944.
22. Hass, G. M. Elastic tissue. *Arch. Path.*, 27: 583, 1939.
23. Thoma. Cited by Sailer, S. [*11*].
24. Costa. Cited by Sailer, S. [*11*].

Nitrogen, Mineral, Uric Acid and Basal Metabolism Studies in a Case of Adult Acute Leukemia with Extensive Osteolytic Bone Disease*

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BONE changes in children with acute leukemia have been described in detail by Silverman [*1*], Dale [*2*], Karpinski [*3*] and others [*4–8*], and bone lesions in adults with chronic leukemia have been recorded by others [*9–11*]. Roentgenographic manifestations of skeletal lesions in adult acute leukemia, however, have been reported infrequently. Kostichek [*12*] described a case of acute monocytic leukemia with diffuse bone involvement. Doub and Hartman [*13*] reported similar findings in acute myelogenous leukemia.

This report concerns an adult with acute lymphocytic leukemia accompanied by extensive involvement of the bones. The balances of nitrogen, potassium, phosphorus, calcium, sodium and chloride, the urinary excretion of uric acid, and the basal metabolic rate were measured before and during chemotherapy. Theoretical balances [*20*] of calcium, nitrogen and potassium based primarily on phosphorus balance were computed. Discrepancies between actual and theoretical balances have been critically inspected for compatibility with hypothetical abnormalities in nitrogen and potassium metabolism in acute leukemia before and during chemotherapy.

CASE REPORT

A forty-six year old white unmarried woman was admitted to the Clinical Center with the chief complaint of "weakness" of five months' duration.

The patient had been well until five months prior to admission when she noted increasing fatigue and weakness. Four months prior to admission, clinical and laboratory examinations were within normal limits except for a differential white blood cell count which showed 80 per cent lymphocytes of varying degrees of maturity. A diagnosis was not established. Two months prior to admission the patient experienced severe, lancinating lumbosacral pain which radiated down the lateral aspect of the left thigh to the knee and pain across the metatarsal arch upon walking. One month prior to admission the patient was again seen by her physician who found no hepatosplenomegaly or lymphadenopathy. Films of the lumbosacral spine revealed no abnormalities. Roentgenograms of both feet revealed demineralization of the head of the fourth metatarsal on the left foot and of the third and fourth metatarsals on the right foot. Blood studies indicated hemoglobin of 13 gm. per 100 ml.; red blood cell count, 4,600,000 per cu. mm.; platelets, 194,000 per cu. mm.; and white blood cell count, 17,850 per cu. mm. The differential count showed 14 per cent granulocytic neutrophils, 47 per cent mature lymphocytes and 39 per cent abnormal immature lymphocytes. Two weeks prior to admission bone marrow biopsy specimens and sternal marrow aspiration specimens were obtained, and a diagnosis of acute leukemia was made. Except for a blood transfusion one week prior to admission, the patient had received no therapy. Symptoms of infection, fever and hemorrhagic phenomena were not significant in the present illness.

The patient had had infectious hepatitis in 1935 and a duodenal ulcer which responded to a medical regimen in 1946. The remainder of the past history, review of systems and family history were non-contributory.

On admission the patient appeared to be a well developed, well nourished white woman, chronically ill, but in no acute distress. Her vital signs were within normal limits. There was no evidence of bleeding in the skin, mucous membranes or fundi. There was no

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significant enlargement of any of the lymph node groups. The liver edge was palpable 5 cm. below the right costal margin and the spleen tip 2 cm. below the left costal margin. Neither organ was tender. There was percussion tenderness over the lower lumbar and sacral vertebrae. There was moderate pain on palpation of the left and right metatarsophalangeal joints but no evidence of inflammation. Neurological examination was normal.

A summary of the laboratory data obtained on admission follows: Urinalysis, blood urea nitrogen, cholesterol, electrolytes, bilirubin, total protein, albumin, globulin, fasting blood sugar and electrocardiogram were within normal limits. Electrophoretic analysis of the patient's serum proteins showed slight elevations in alpha-2 and gamma globulin with an albumin at the lower range of normal. The hemoglobin was 11.0 gm. per 100 ml.; hematocrit, 33.5 per cent; platelets, 105,000 per cu. mm.; white blood cell count, 21,400 per cu. mm. Differential count showed 5 segmented neutrophils, 31 lymphocytes, 2 monocytes and 63 abnormal immature lymphocytes ("lymphoblasts"). A diagnosis of acute lymphocytic leukemia was confirmed after examination of supravital and fixed preparations of a bone marrow aspiration.

X-ray examination showed progression of the destructive lesions involving the third and fourth left metatarsal heads and the third right metatarsal head. Punctate destruction of the calvarium was most prominent in the anteroparietal and frontal bones. Films of the pelvis revealed numerous minute punched-out areas of decreased density in the ilium and proximal ends of the femora. Other films taken on admission showed marked osteoporosis of the cervical, dorsal and lumbar spine and rib cage.

At the beginning of her third hospital day, the patient was placed on a metabolic study regimen. During the first two weeks of hospitalization (control phase, periods 1 and 2) the white blood cell count fell from 21,400 to 11,000 per cu. mm. without therapy. On the fourteenth hospital day chemotherapy consisting of 2.5 mg. of 4-amino-N^{10} methyl pteroyl glutamic acid (methotrexate) and 250 mg. of 6-mercaptopurine (purinethol®) daily was begun. Although serial bone marrow aspirations subsequently demonstrated evidence of relative increase in granulopoiesis, thrombopoiesis and lymphocytic maturation, the peripheral counts revealed persistent leukopenia, thrombocytopenia and anemia.

The patient's course was marked by numerous complications. On the tenth hospital day an unexplained fever developed. On the fifteenth hospital day penicillin and streptomycin were administered and continued until the twenty-eighth hospital day. The fever subsided on the seventeenth day. She continued to complain of lancinating lumbar pain radiating to the backs of both legs. Anorexia developed, and she began to refuse an occasional feeding. On the twenty-sixth hospital day excruciatingly painful muscle spasms developed involving the hamstring groups. Failure of narcotics to relieve back pain led to a trial of palliative radiotherapy, which was followed by relief of symptoms. On the twenty-sixth day anemia of 5.2 gm. hemoglobin per 100 ml. necessitated a transfusion of 500 ml. whole blood which raised the hemoglobin to 7.9 gm. per 100 ml. a level maintained for ten days.

On the thirtieth hospital day, the patient complained of a sore tongue. It appeared smooth and erythematous, but lingual and buccal ulcerations characteristic of methotrexate toxicity did not develop. On the thirty-fifth hospital day culture of the oral cavity revealed Candida. One per cent gentian violet was applied locally. Large doses of B-complex vitamins were administered.

By the thirty-third hospital day the white blood cell count had risen from a low of 400 to 1,025 per cu. mm., and the platelets had reached the 100,000 per cu. mm. range. These counts were maintained until the thirty-seventh day when they began to fall. Electrophoretic studies of the patient's serum proteins showed a slight drop in total protein and in alpha-2 and gamma globulin with a slight rise in albumin and alpha-1 globulin. Another 500 ml. of whole blood given on the thirty-sixth day raised the hemoglobin level to 9.4 gm. per 100 ml.

On the thirty-eighth hospital day an unexplained fever again developed which persisted between 38.4° and 39.4°C. until the patient's death. Streptomycin and penicillin therapy was reinstituted. She had one copious stool late in the evening followed by two liquid stools the next day. She vomited bile-stained material at frequent intervals. On the fortieth day administration of methotrexate and 6-mercaptopurine was discontinued. By the forty-second day the patient was icteric. Her liver was very tender and palpable 10 cm. below the costal margin. Liver function studies revealed cephalin flocculation test, 3 plus; thymol turbidity test, 1 plus; total bilirubin, 4.8 mg. per cent; one-minute bilirubin, 2.4 mg. per cent; and alkaline phosphatase, 2.6 Bessey, Lowry and Brock units. Bile was present in the urine but urine urobilinogen was not increased. The metabolic regimen was discontinued at 7:00 A.M. on the forty-third hospital day.

During the forty-third day the patient's condition was critical. The blood urea nitrogen was 155 mg. per cent; sodium 122 mEq./L.; potassium, 4.1 mEq./L.; and carbon dioxide content 21 mEq./L. The possibility that an acute hepatitis was responsible for the progressively downhill course led to the administration of large intravenous doses of hydrocortisone. Her blood pressure fell and remained at hypotensive levels despite levophed.® She died quietly shortly after midnight on the forty-fourth hospital day.

Postmortem films showed progression of the osteolytic process in the third and fourth left metatarsal

heads. Multiple small areas of destruction of all the bony structures of the foot were also visible. Films of the skull and long bones demonstrated progressive osteolysis.

An autopsy was performed nine hours postmortem. A summary of significant findings follows: The sclerae and skin were lemon yellow. The esophagus was ulcerated throughout its entire length. The first portion of the jejunum appeared gangrenous, although no gross perforations were seen. Two sessile yellow polyps were located in the distal jejunum. The spleen weighed 125 gm. and the liver weighed 2,200 gm. Both adrenal glands were enlarged, the left weighed 8.9 gm. and the right 7.6 gm. The right and left kidney weighed 120 and 160 gm., respectively. All samples of marrow were soft, tan and aplastic.

Microscopic examination of the tongue revealed a severe uniform ulceration of the surface epithelium with a diffuse superficial mantle of fungus growth proved to be Candida albicans. Extensive ulcerations of the mucosa of the esophagus and jejunum were overgrown with C. albicans. The underlying jejunal mucosa was necrotic. Carcinoid tumors and a membrane of bacterial colonies and C. albicans were found in the ileum. Sections of the liver revealed congestion of the sinusoids and vacuolated parenchymal cells containing brown and orange pigment. No leukemic infiltrates were seen. Fat stains demonstrated large sudanophilic droplets in the parenchymal cells at the periphery of the lobules. There was bile stasis in the canaliculi and small bile ducts. Sections of the kidneys revealed swollen, congested glomeruli and protein material in the intracapsular space. The proximal convoluted tubules, Henle's loops, and the distal convoluted tubules contained protein precipitates, highly refractile deposits and some hyalin casts.

Sections of bone showed severe atrophy of the marrow with hemorrhage and in some areas granulation tissue. Severe osteoporosis was evident in all bones examined. In some areas numerous lacunae in the cortical and trabecular bone suggested recent osteoclastic activity.

METABOLIC STUDY

METHODS

The balances of nitrogen, potassium, phosphorus, calcium, sodium and chloride, and the urinary excretion of uric acid, were measured during seven periods from the third through the forty-third hospital day. Period 2 was divided (periods 2a and 2b). Urines collected in period 4 were separated into three pools (periods 4a, 4b and 4c). Period 7 consisted of five days while all other periods were six days in duration.

During the entire study the patient was on a metabolic unit under the constant supervision of nurses trained in the technics of metabolic investigation. The diet was prepared in a metabolic kitchen under the direct supervision of a research dietitian. Items of the diet were weighed individually and when cooked were prepared in separate containers. Canned goods from the same lot and trimmed meat from one carcass were used. The daily diet constantly contained 1,297 calories, 59.6 gm. of fat, 144 gm. of carbohydrate, 45.2 gm. of protein, 538 mg. of phosphorus, 114 mg. calcium, 8.8 mEq. of sodium, 6.0 mEq. of chloride and 42.1 mEq. of potassium. Refused food and emesis were carefully saved, analyzed and subtracted from the daily intake for that period. The constituents of blood were added to intake when transfusions were given. A fixed quantity of distilled water for drinking purposes was provided daily.

The patient was weighed each day before breakfast in a tared robe on a Fairbanks-Morse platform scale accurate to 10 gm. Fasting blood samples for chemical determinations were obtained from an antecubital vein without the use of a tourniquet. Blood for hematologic examination was obtained approximately one hour after breakfast.

Voided urine specimens were immediately pooled in a refrigerated jar without preservative. At the end of each twenty-four-hour period an aliquot of the refrigerated specimen was frozen until the analyses were performed. Feces were combined into six-, five- or three-day pools demarcated by carmine markers.

Food and fecal specimens were homogenized in a 5 L. Model CB-2 Waring Commercial Blender. Weighed aliquots of homogenates were used for nitrogen and chloride analyses. Other weighed and tared aliquots were dried by lyophilization. Weighed samples of the pulverized, dry material were ashed in Vycor crucibles at 400°c. prior to analyses for potassium, phosphorus, calcium and sodium.

The following analytical methods were employed: urinary nitrogen, micro-Kjeldahl [*14*]; food and fecal nitrogen, macro-Kjeldahl [*14*]; potassium and sodium, internal standard flame photometer [*15*]; phosphorus, Taussky and Shorr [*16*]; calcium, Kochakian and Fox [*17*]; urine chloride, modified Sendroy [*18*], food and fecal chloride, modification of the method described by Peters and van Slyke [*18*]; uric acid, enzymatic method of Praetorius [*19*].

During periods 1, 2a and 2b (twelve days), the patient received no therapy. During periods 3, 4, 5 and 6 and during the first three days of period 7 (twenty-seven days) she received 6-mercaptopurine and methotrexate. The patient was given a unit of blood (500 ml.) in period 5 and period 6.

RESULTS

Balance data appear in Table 1 and are presented graphically in Figure 1 according to the system of Reifenstein, Albright and Wells [*20*]. Theoretical balances computed using factors compiled for muscle and bone by Reifenstein,

TABLE I

METABOLIC BALANCE DATA, HEMATOLOGY AND BLOOD CHEMISTRIES

Period	Hospital Days	Therapy*	Basal Metabolic Rate (Cal./M²/hr.)	Weight (kg.)	Nitrogen (gm./24 hr.)				Potassium (mEq./24 hr.)				Phosphorus (mg./24 hr.)			
					Intake	Feces	Urine	Balance	Intake	Feces	Urine	Balance	Intake	Feces	Urine	Balance
1	3–8	None	46.0	46.11	7.22	1.03	8.34	−2.15	42.1	9.8	39.4	−7.1	538	336	604	−402
2A	9–11	None	50.4	45.56	7.22	0.80	8.22	−1.80	42.1	6.7	47.0	−11.6	538	256	558	−276
2B	12–14	None	57.9	45.35	7.22	1.06	9.82	−3.66	42.1	9.6	58.8	−23.3	538	396	713	−571
2 Tot.	9–14	None	....	45.45	7.22	0.93	9.02	−2.73	42.1	8.2	51.4	−17.5	538	326	635	−432
3	15–20	CC	50.8	44.66	7.22	1.18	9.27	−3.23	42.1	18.0	41.0	−16.9	538	460	1069	−991
4A	21–22	CC	....	43.78	5.03	0.94	6.10	−2.01	33.7	12.6	30.4	−9.3	453	327	680	−554
4B	23–24	CC	....	44.46	7.22	0.94	10.51	−4.23	42.1	12.6	59.4	−29.9	538	327	1061	−850
4C	25–26	CC	....	44.29	4.01	0.94	7.68	−4.61	29.8	12.6	51.5	−34.3	417	327	936	−846
4 Tot.	21–26	CC	....	44.18	5.42	0.94	8.10	−3.62	35.2	12.6	47.1	−24.5	469	327	893	−751
5	27–32	CC-R	39.6	42.79	5.02	0.50	5.92	−1.40	21.1	6.3	31.4	−16.6	279	178	369	−268
6	33–38	CC-R-V	39.6	41.61	7.15	0.78	5.75	−0.62	17.7	9.3	28.5	−20.1	287	324	443	−480
7	39–43	CC-R-V	50.2	39.60	4.75	1.38	3.73	−0.36	17.7	17.4	10.5	−10.2	273	217	283	−227

Period	Calcium (mg./24 hr.)				Sodium (mEq./24 hr.)				Chloride (mEq./24 hr.)				Urine Uric Acid (gm./24 hr.)	Serum Uric Acid (mg. %)	White Blood Cells (10^3 per cu. mm.)	Hemoglobin (gm. %)
	Intake	Feces	Urine	Balance	Intake	Feces	Urine	Balance	Intake	Feces	Urine	Balance				
1	114	315	241	−442	8.8	0.2	21.7	−13.1	5.96	0.08	19.44	−13.6	0.870	7.2	19.9	11.3
2A	114	201	223	−310	8.8	0.1	5.7	+2.8	5.96	0.15	7.8	−2.0	0.819	...	17.5	9.9
2B	114	274	160	−320	8.8	0.8	6.6	+1.4	5.96	0.81	10.2	−5.0	1.170	7.3	.....	9.5
2 Tot.	114	238	192	−316	8.8	0.4	6.2	+2.2	5.96	0.48	9.0	−3.5	0.995	...	.....	....
3	114	333	102	−321	8.8	2.4	0.8	+5.6	5.96	1.65	2.4	+1.9	1.507	7.2	1.02	7.7
4A	100	224	82	−206	7.4	0.3	0.2	+6.9	4.18	0.21	2.0	+2.0	0.994	5.4	.98	7.2
4B	114	224	68	−178	8.8	0.3	0.3	+8.2	5.96	0.21	7.8	−2.0	1.148	...	.62	5.8
4C	96	224	26	−154	7.8	0.3	0.3	+7.2	4.70	0.21	6.0	−1.6	0.896	...	.45	5.2
4 Tot.	103	224	59	−180	8.0	0.3	0.3	+7.4	4.95	0.21	5.3	−0.6	1.013	...	.....	.....
5	60	171	20	−131	14.6	0.5	0.2	+13.9	10.1	0.21	1.2	+8.7	0.482	5.0	.40	7.4†
6	53	317	6	−270	8.7	9.0	0.2	−0.5	6.4	0.24	0.8	+5.4	0.248	3.8	.96	7.4
7	−63	400	2	−465	−16.5	18.7	0.3	−35.3	−31.47	10.74	0.2	−42.4	0.113	...	.21	8.6†

* CC—combined chemotherapy.
R—raudixin.®
V—vitamins.
† After transfusion.

Albright and Wells [*20*] and for leukemic tissue by Waterhouse, Terepka and Sherman [*21*] are presented in Table II.

A "theoretical balance" is a derived value representing the net gain (if positive) or loss (if negative) of one element which should occur simultaneously with the measured values of other elements. For example, a theoretical nitrogen balance can be derived from the measured balances of phosphorus and calcium assuming that these three elements bear a fixed ratio to one another in a given tissue and that this tissue is being synthesized (anabolism) or destroyed (catabolism). The ratios nitrogen/phosphorus (corrected for calcium), potassium/nitrogen, and potassium/phosphorus (corrected for calcium) for normal muscle [*20*] and for leukemic tissue [*21*], the calcium/phosphorus ratio for bone and the sodium/chloride ratio for extracellular fluid [*20*] have been published. All phosphorus balances are corrected for the phosphorus accompanying calcium by using the factor 2.23. From these ratios the theoretical balances which should have occurred in this study have been calculated and compared with the actual balances observed. Discrepancies between theoretical and observed values require explanation and lead to interpretations of data to be cited. The ratios on which theoretical balances are based were computed from relatively few original observations, are highly variable among tissues and even within the same tissue, do not reflect over-all body composition and include all experimental error. Therefore, interpretation must be made and viewed with some degree of caution.

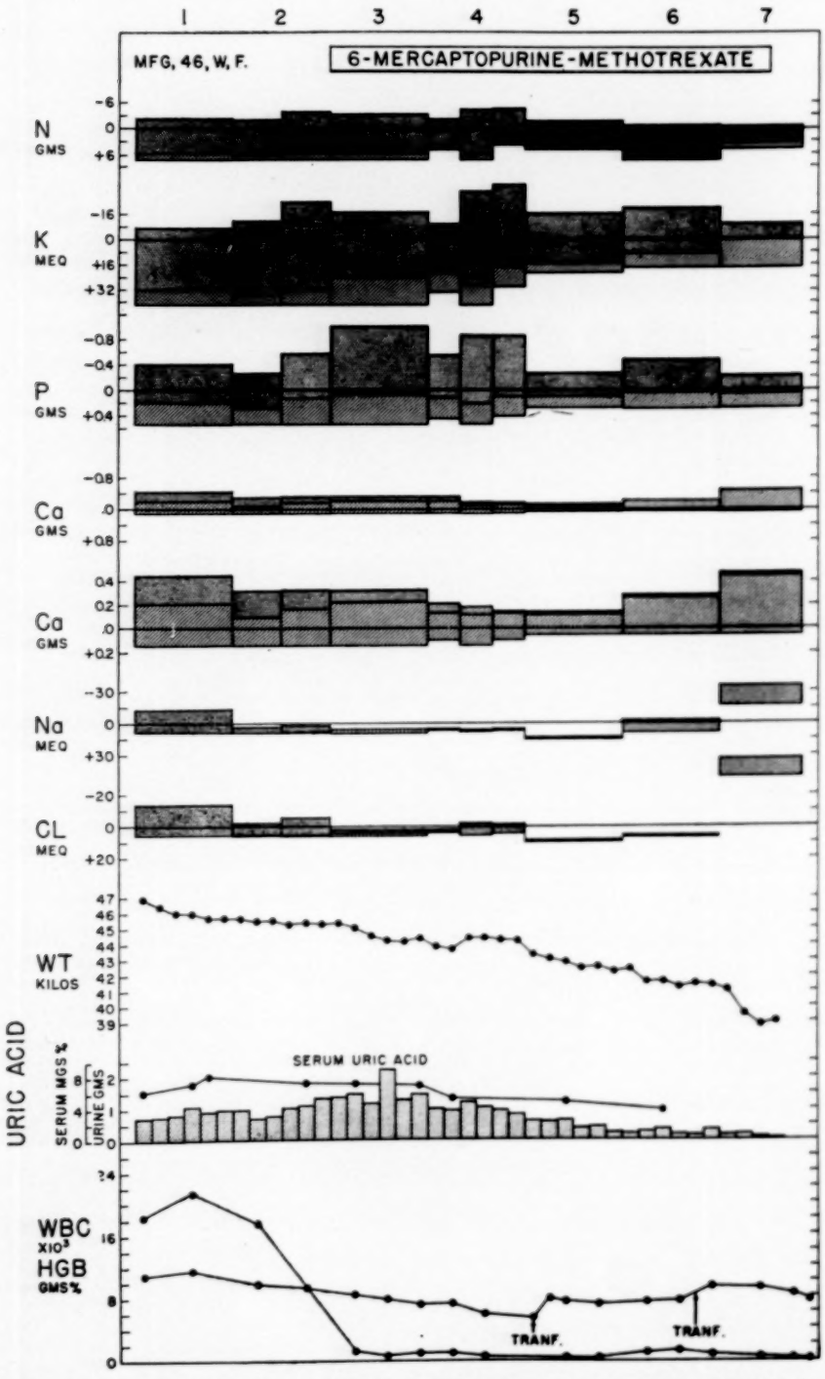

FIG. 1. Metabolic balances, body weight, uric acid excretion and hematologic data before and during combined chemotherapy. The balance charts are constructed by plotting intake downward from the zero line and then plotting upward from the intake line, first fecal excretion and second urinary excretion. If the sum total of excretion coincides with the zero line, equilibrium is indicated; if it extends above, the balance is negative; and if it lies below, the balance is positive. The ordinates of the nitrogen, potassium, phosphorus and the first of the calcium charts are so selected that equal heights represent the approximate ratios at which these elements exist in normal protoplasm and bone, i.e., 3 gm. of nitrogen to 8 mEq. of potassium to 200 mg. of phosphorus (protoplasm) and 400 mg. of calcium to 200 mg. of phosphorus (bone). The second calcium chart is identical to the first except that the scale has been expanded approximately three times to provide easier reading. The Arabic numerals along the top of the chart refer to balance periods of the study and may be identified with hospital days by reference to Table 1.

CALCIUM BALANCE

During the twelve-day control phase, calcium balance was negative, averaging −379 mg. per twenty-four hours (or approximately −8.7 mg. per kg. per twenty-four hours). Urinary calcium, while averaging 216 mg. per twenty-four hours, decreased from 251 to 122 mg. per twenty-four hours during the control phase. Theoretical calcium balances based on phosphorus balance (corrected for nitrogen) were computed twice, once using the nitrogen/phosphorus ratio for muscle (14.7) and again using the nitrogen/phosphorus ratio for leukemic tissue (7.3). During the control phase, theoretical values based on the muscle ratio (14.7) averaged −534 mg. per twenty-four hours and exceeded actual calcium losses by an average of 155 mg. per twenty-four hours. Theoretical values based on the leukemic tissue ratio (7.3) averaged 174 mg. per twenty-four hours and were less than actual calcium losses by an average of 205 mg. per twenty-four hours.

The large negative balance of calcium is compatible with extensive osteolytic bone disease [*22–25*]. The magnitude of this negative balance is too great to be attributed to the low calcium intake [*25–27*]. The gradual decrease in urinary calcium, however, does suggest either a spontaneous remission of disease activity or some degree of physiologic adaptation of the low calcium diet [*25*]. The discrepancies between the actual balance and the theoretical balance suggest that the non-bony tissue being destroyed was in all probability a mixture of muscle and leukemic tissue.

During the initial six days of the treatment phase (period 3), calcium balance remained unchanged at −321 mg. per twenty-four hours. Urinary calcium continued to decrease to an average of 102 mg. per twenty-four hours. Theoretical calcium balances, however, averaged −1,713 and −1,220 mg. per twenty-four hours and exceeded actual calcium balances by 1,392 mg. per twenty-four hours and 899 mg. per twenty-four hours when computed on the muscle and leukemic tissue ratios, respectively. These marked increases in theoretical calcium losses were attributed to huge increases in phosphorus losses unaccompanied by nitrogen.

During periods 4 and 5 (seventh through the eighteenth day of treatment) calcium balance decreased to an average of −131 mg. per twenty-four hours. Urinary calcium fell further to an average of 20 mg. per twenty-four hours. In

TABLE II

ACTUAL BALANCES, THEORETICAL BALANCES AND BALANCE RATIOS

Period	Actual Balances					Theoretical Balances						Balance Ratios			
	N (gm./24 hr.)	K (mEq./24 hr.)	P_α (gm./24 hr.)	Ca (gm./24 hr.)	Na (mEq./24 hr.)	Cl (mEq./24 hr.)	$P_\beta = Ca/2.23$ (gm./24 hr.)	$P_\alpha - P_\beta$ (gm./24 hr.)	$N_\mu = P_{\alpha-\beta} \times 14.7$ (gm./24 hr.)	$N_\lambda = P_{\alpha-\beta} \times 7.3$ (gm./24 hr.)	$K_\mu = P_{\alpha-\beta} \times 40$ (mEq./24 hr.)	$N_\lambda = P_{\alpha-\beta} \times 30$ (mEq./24 hr.)	$N_\mu = K/2.7$ (gm./24 hr.)	$N_\lambda = K/4.2$ (gm./24 hr.)	$Na = Cl \times 1.5$ (mEq./24 hr.)	$N/P_{\alpha-\beta}$	K/N	$K/P_{\alpha-\beta}$	Na/Cl
1	−2.15	−7.1	−.402	−.442	−13.1	−13.6	−.198	−.204	−3.00	−1.49	−8.2	−6.1	−2.63	−1.69	−20.4	10.5	3.3	34.8	1.0
2a	−1.80	−11.6	−.276	−.310	+2.8	−2.0	−.139	−.137	−2.01	−1.00	−5.5	−4.1	−4.30	−2.76	−3.0	13.1	6.4	84.8	−1.4
2b	−3.66	−23.3	−.571	−.320	+1.4	−5.0	−.143	−.428	−6.29	−3.12	−17.1	−12.8	−8.63	−5.55	−7.5	8.6	6.4	54.5	−2.8
2 Total	−2.73	−17.5	−.423	−.316	+2.2	−3.5	−.142	−.281	−4.13	−2.05	−11.2	−8.4	−6.48	−4.17	−5.3	9.7	6.4	62.3	−6.3
3	−3.23	−16.9	−.991	−.321	+5.6	+1.9	−.144	−.847	−12.45	−6.18	−33.9	−25.4	−6.26	−4.02	+2.8	3.8	5.2	19.9	+2.9
4a	−2.01	−9.3	−.554	−.206	+6.9	+2.0	−.092	−.462	−6.79	−3.37	−18.5	−13.9	−3.44	−2.21	+3.0	4.4	4.6	20.1	+3.4
4b	−4.23	−29.9	−.850	−.178	+8.2	−2.0	−.080	−.770	−11.32	−5.62	−30.8	−23.1	−11.07	−7.12	−3.0	5.5	7.1	38.9	−4.1
4c	−4.61	−34.3	−.846	−.154	+7.2	−1.6	−.069	−.777	−11.42	−5.67	−31.1	−23.3	−12.07	−8.17	−2.4	5.9	7.4	44.2	−4.5
4 Total	−3.62	−24.5	−.751	−.180	+7.4	−0.6	−.081	−.670	−9.85	−4.89	−26.8	−20.1	−9.07	−5.83	−0.9	5.4	6.8	36.6	−12.3
5	−1.40	−16.6	−.268	−.131	+13.9	+8.7	−.059	−.209	−3.07	−1.53	−8.4	−6.3	−6.15	−3.95	−13.0	6.7	11.9	79.5	+1.6
6	−0.62	−20.1	−.480	−.270	−0.5	+5.4	−.121	−.359	−5.28	−2.62	−14.4	−10.8	−7.44	−4.79	+8.1	1.7	32.4	56.0	−0.1
7	−0.36	−10.2	−.227	−.465	−35.3	−42.4	−.209	−.018	−0.26	−0.13	−0.7	−0.5	−3.78	−2.43	−63.6	2.0	28.4	567	+0.8

NOTE: P_α = actual phosphorus balance.
P_β = phosphorus associated with calcium in bone (Ca/2.23).
$P_{\alpha-\beta}$ = phosphorus balance corrected for calcium.
N_μ = theoretical nitrogen balance based on ratios for muscle ($P_{\alpha-\beta} \times 14.7$; K/2.7).
N_λ = theoretical nitrogen balance based on ratios for leukemic tissue ($P_{\alpha-\beta} \times 7.3$; K/4.2).
K_μ = theoretical potassium balance based on ratios for muscle ($P_{\alpha-\beta} \times 40$).
K_λ = theoretical potassium balance based on ratios for leukemic tissue ($P_{\alpha-\beta} \times 30$).

period 4 theoretical negative calcium balances remained large (−1,125 mg. and −567 mg. per twenty-four hours) and were far greater (−945 mg. and −387 mg. per twenty-four hours) than actual balances. In period 5, theoretical negative calcium balances became smaller (−386 mg. and −169 mg. per twenty-four hours) and the excess over actual calcium balance was reduced (−255 and −38 mg. per twenty-four hours).

The decrease in negative calcium balances and the decrease in urinary calcium in periods 4 and 5 could be interpreted as a favorable effect of the antimetabolites on the course of the bone disease. However, since urinary calcium had begun to fall during the control periods, these changes may have been unrelated to antimetabolite therapy. Spontaneous changes in disease activity or a gradual adaptation to a low calcium diet offer other interpretations. Decreases in theoretical calcium balances resulted from the marked drop in the loss of phosphorus during period 5.

In periods 6 and 7, marked increases in negative calcium balances were observed. Urinary calcium continued to fall to an average of 2 mg. per twenty-four hours. The increased calcium loss occurred entirely in the feces. Theoretical calcium losses rose in period 6 in association with a rise in urinary phosphorus but fell again in period 7. The excess of theoretical balance over actual calcium balance was negligible in period 7.

Negative calcium balances approaching control levels in periods 6 and 7 suggest renewed bone destruction by the leukemic process. Fecal excretion of almost all calcium may be attributed to diarrhea and to deteriorating renal function.

NITROGEN AND PHOSPHORUS BALANCES

During the control phase, nitrogen balance was negative, averaging −2.44 gm. per twenty-four hours. Phosphorus balance (corrected for calcium) averaged −.242 gm. per twenty-four hours. Theoretical nitrogen balance based on phosphorus balance (corrected for calcium) and the nitrogen/phosphorus ratio for muscle (14.7) indicate that nitrogen was retained in excess of theoretical nitrogen balance. However, using the nitrogen/phosphorus ratio (7.3) for leukemic cells, nitrogen was lost in excess of theoretical nitrogen balance. These observations, can be interpreted as indicating simultaneous destruction of both normal and leukemic tissue during the control period.

In periods 3 and 4, during the treatment phase, nitrogen balances averaged −3.42 gm. per twenty-four hours. Phosphorus balances (corrected for calcium) averaged −.758 gm. per twenty-four hours. Theoretical nitrogen balances based on phosphorus balances (corrected for calcium) and the nitrogen/phosphorus ratio for leukemic tissue indicate that large amounts of nitrogen were being retained in excess of theoretical nitrogen balances.

During periods 5 and 6 nitrogen balances were less negative, averaging −1.01 gm. per twenty-four hours. Phosphorus balances (corrected for calcium) were −.334 gm. per twenty-four hours. Theoretical nitrogen balances based on phosphorus balance and the nitrogen/phosphorus ratio for leukemia tissue again indicate that nitrogen was being retained in excess of phosphorus.

In the final period, chemotherapy was administered for only three days. Nitrogen and phosphorus (corrected for calcium) balances were close to equilibrium (−0.36 and −.018 gm. per twenty-four hours). Theoretical nitrogen balance approximated actual balance.

The retention of nitrogen throughout the treatment phase, in excess of that attributed to phosphorus, and the nitrogen/phosphorus ratio for leukemic tissue suggest selective retention of nitrogen without phosphorus either by the host or by the tumor.

POTASSIUM BALANCE

Potassium balances averaged −12.3 mEq. per twenty-four hours during the control phase, −20.7 mEq. per twenty-four hours in treatment periods 3 and 4, −18.3 mEq. per twenty-four hours in treatment periods 5 and 6, and −10.3 mEq. per twenty-four hours in period 7. The possibility of nitrogen retention by host or tumor is supported also by the high potassium/nitrogen balance ratios (6.4 to 32.4) in periods 2 through 7. These high balance ratios may also reflect an exchange of intracellular potassium for extracellular sodium. The theoretical potassium balance based on phosphorus (corrected for calcium) and the potassium/phosphorus ratio for muscle exceeded the observed balances in control period 1 but not in control period 2. Control phase potassium losses were always greater than the theoretical losses based on phosphorus balance (corrected for calcium) and the potassium/phosphorus ratio for leukemic tissue. In treatment periods 3 and 4a, the observed potassium losses were less than those theoretically based on phosphorus balance (corrected for calcium) and

the potassium/phosphorus ratio for leukemic tissue. In periods 4b through 7, the observed potassium loss always exceeded the theoretical loss based on phosphorus balance (corrected for calcium) and the potassium/phosphorus ratio for leukemic tissue. Except in period 4b the observed potassium loss also exceeded the theoretical loss based on the potassium/phosphorus ratio for muscle tissue.

Theoretical potassium balances may be interpreted as indicating a shift of potassium from intra- to extracellular fluid in control phase period 2 and in treatment periods 4c through 7, since even destruction of muscle tissue could not account for all the potassium lost.

Theoretical nitrogen balances based on potassium balances and the potassium/nitrogen ratio for leukemic tissue exceeded observed nitrogen losses in every period except period 1 when it exceeded that theoretically based on potassium balance and the potassium/nitrogen ratio for muscle.

These discrepancies between actual and theoretical nitrogen balances again suggest selective retention of nitrogen by the host or the tumor. In addition, they may also reflect the loss of intracellular potassium in exchange for sodium. This latter possibility is compatible with the development of hyponatremia, the high ratio of observed to theoretical potassium loss based on phosphorus (corrected for calcium), and the positive sodium balances.

SODIUM AND CHLORIDE BALANCES

Sodium balances were positive or were in equilibrium except in period 1 when the patient was adapting to the low salt diet and in period 7 when she vomited and had diarrhea. Chloride balances were slightly more negative or less positive than sodium balances except in period 6. Actual sodium balance was more positive or less negative than theoretical sodium balance based on chloride balance and a sodium/chloride ratio of 1.5 [*20*] except in period 6. Sodium and chloride balances suggest an exchange of intracellular potassium for extracellular sodium. The negative balances of sodium and chloride in periods 1 and 7 were associated with a marked decrease in weight suggestive of substantial losses of extracellular fluid.

URIC ACID

Serum uric acid levels of 6 to 8 mg. per cent and urinary uric acid excretions averaging 932 mg. per twenty-four hours during the control phase indicate increased uric acid production. The temporary increment in urinary uric acid to 1507 mg. per twenty-four hours during the first treatment period (period 3) implies tissue destruction, as does the increment in phosphorus loss. However, the simultaneous decline in serum and urinary uric acid suggests that one of the agents used in chemotherapy may have interfered with the production of uric acid so that its availability for excretion became negligible.

BASAL METABOLIC RATE

Basal metabolic rate was determined on eight occasions by experienced technicians using the Sanborn "Metabolizer." Four determinations during the control phase averaged 50.1 calories per square meter per hour or +39.2 per cent by the Aub-Dubois standards. A protein-bound iodine determination [*29*] during control period 1 was 6.2 μg. per 100 ml., and a determination of radioactive iodine uptake [*30*] was 42 per cent in twenty-four hours. At the end of the first six days of treatment, the patient's basal metabolic rate was 50.8 calories per square meter per hour, or +41.1 per cent by the Aub-Dubois standards. At the beginning of period 5 the patient was given 100 mg. of raudixin® twice daily orally. Determinations of basal metabolic rate in period 5 after one day of raudixin therapy and in period 6 after eight days of raudixin therapy were identical, 39.6 calories per square meter per hour or +10 per cent by the Aub-Dubois standards. Administration of raudixin was discontinued on the second day of period 7. Two days later the basal metabolic rate was 50.2 calories per square meter per hour or +39.5 per cent by Aub-Dubois standards. These observations indicate an elevated metabolic rate not attributable to hyperthyroidism and not altered by combined chemotherapy. The marked lowering of basal metabolic rate coincident with administration of the tranquilizing agent raudixin, together with the return to high levels on withdrawal of the drug, strongly suggest that the patient's anxiety contributed to the elevated metabolic rate. However, a direct effect of the drug on the metabolic rate is an alternative possibility. It is noteworthy too that the basal metabolic rate even with the administration of raudixin was still elevated, a finding compatible with the theory that malignancy is associated with an increase in metabolic rate.

COMMENTS

From the foregoing computations it is apparent that the balance ratios nitrogen/phosphorus (corrected for calcium), potassium/nitrogen and potassium/phosphorus (corrected for calcium) were appreciably different from values based on the composition of muscle or of leukemic tissue. It is also clear that two factors, a selective retention of nitrogen and a shift of potassium from the cells, could explain the discrepancies between observed and theoretical balances of calcium, nitrogen and potassium.

The cumulative nitrogen theoretically retained on the basis of the nitrogen/phosphorus ratio for muscle during the seven periods of the study amounts to 94.9 gm., or enough to form almost 3 kg. of protoplasm. The site of nitrogen retention of this magnitude is uncertain, although theoretically an 800 gm. increase in the size of the liver and a 150 mg. per cent increase in non-protein nitrogen would account for 90 per cent of the total. The cause of this nitrogen retention is not clear. It is obvious, however, that it was accelerated dramatically by combined chemotherapy. Heaney and Eliel [*28*] made similar observations in a study of a three year old girl with acute leukemia who was treated by 6-mercaptopurine alone. They attributed their findings to destruction of leukemic tissue rich in phosphorus and to an increase in normal tissue anabolism due to the removal of competition or inhibition by the destruction of neoplastic tissue. In view of the many differences between these two situations, no direct comparison is possible. However, as already mentioned, in this adult patient there was no indication that normal or accelerated muscle anabolism occurred. These observations are more compatible with the suggestion that nitrogen retention and potassium loss may be undesirable side effects, hitherto undescribed, of combined chemotherapy.

CONCLUSIONS

Acute leukemia in this patient resulted in the destruction of both normal soft tissue and bone. In addition, destruction of a tissue rich in phosphorus, presumably leukemic cells, occurred simultaneously and was accelerated by chemotherapy. This destruction was reflected by high control serum and urinary uric acid levels and by a transient rise in uric acid excretion with chemotherapy. The subsequent fall in both serum and urinary uric acid levels suggests a decreased rate of tissue destruction and also, in view of the extremely low levels attained, inhibition of uric acid production by chemotherapy. As leukemic cells and host protoplasm were destroyed, phosphorus and potassium were excreted but nitrogen was selectively retained. The hepatomegaly observed at autopsy affords one possible site for nitrogen deposition. Non-protein nitrogen elevation affords another. Exchange of intracellular potassium for sodium occurred as the patient's condition deteriorated. Bone destruction was inhibited transiently if at all by chemotherapy.

SUMMARY

A metabolic study in an adult case of acute lymphocytic leukemia with extensive osteolytic bone disease is reported. During a twelve-day control phase, bone, normal soft tissue and presumably leukemic tissue were destroyed at a rapid rate. Phosphorus and uric acid excretion rose with the institution of combined 6-mercaptopurine and methotrexate therapy. Theoretical balances derived from actual balance data suggest that nitrogen was being selectively retained, possibly to produce the organomegaly noted at autopsy unassociated with leukemic infiltration. Exchange of intracellular potassium for extracellular sodium occurred as the patient's condition deteriorated. Bone destruction was transiently inhibited by chemotherapy. Administration of rauwolfia serpentina (raudixin) was associated with a marked reduction in basal metabolic rate.

REFERENCES

1. Silverman, F. N. The skeletal lesions in leukemia. Clinical and roentgenographic observations in 103 infants and children with a review of the literature. *Am. J. Roentgenol.*, 59: 819, 1948.
2. Dale, J. H. Leukemia in childhood, a clinical and roentgenographic study of 72 cases. *J. Pediat.*, 34: 421, 1949.
3. Karpinski, F. E. and Martin, J. F. The skeletal lesions of leukemic children treated with aminopterin. *J. Pediat.*, 37: 208, 1950.
4. Kalayjian, B., Herbut, P. and Erf, L. Bone changes of leukemia in children. *Radiology*, 47: 223, 1946.
5. Clark, J. J. Unusual bone changes in leukemia. *Radiology*, 26: 237, 1936.
6. Jaffe, H. L. Skeletal manifestations of leukemia and malignant lymphoma. *Bull. Hosp. Joint Dis.*, 13: 217, 1952.
7. Baty, J. M. and Vogt, E. C. Bone changes of leukemia in children. *Am. J. Roentgenol.*, 34: 310, 1935.

8. Dresner, E. The bone and joint lesions in acute leukemia and their response to folic acid antagonists. *Quart. J. Med.*, 7o: 339, 1950.
9. Mendl, K. and Saxe, O. Bone changes in leukemia. *Am. J. Roentgenol.*, 44: 31, 1940.
10. Mazet, R. Vertebral manifestations of malignant lymphoma, myeloid leukemia, and multiple myeloma. *Surgery*, 29: 545, 1951.
11. Craver, L. F. and Copeland, M. M. Changes of the bones in the leukemias. *Arch. Surg.*, 30: 639, 1935.
12. Kositchek, R. J. Monocytic leukemia associated with bone changes. *Ann. Int. Med.*, 19: 1008, 1943.
13. Doub, H. P. and Hartman, F. W. Lymphocytic, myelocytic, and monocytic neoplasms. *J. A. M. A.*, 105: 942, 1935.
14. Pierce, C. P. and Hainisch, E. L. Quantitative Analysis, p. 140. New York, 1950. John Wiley & Sons, Inc.
15. Bowman, R. L. and Berliner, R. W. Principles of design and operation of the internal standard flame photometer for sodium and potassium determination. *Federation Proc.*, 8: 14, 1949.
16. Taussky, H. H. and Shorr, E. A microcolorimetric method for the determination of inorganic phosphorus. *J. Biol. Chem.*, 202: 675, 1953.
17. Kochakian, C. D. and Fox, R. P. Microdetermination of calcium by titration of the oxalate with ammonium hexanitratocerate. *Anal. Chem.*, 16: 762, 1944.
18. Peters, J. P. and van Slyke, D. D. Quantitative Clinical Chemistry Methods, vol. 2, pp. 833–835. Baltimore, 1932. Williams & Wilkins Co.
19. Praetorius, E. An enzymatic method for the determination of uric acid by ultraviolet spectrophotometry. *Scandinav. J. Clin. & Lab. Investigation*, 1: 222, 1949.
20. Reifenstein, E. C., Albright, F. and Wells, S. L. The accumulation, interpretation and presentation of data pertaining to metabolic balances, notably those of calcium, phosphorus and nitrogen. *J. Clin. Endocrinol.*, 5: 367, 1945.
21. Waterhouse, C. A., Terepka, A. R. and Sherman, C. D., Jr. The gross electrolyte composition of certain human malignant tissues. *Cancer Research*, 15: 544, 1955.
22. Bartter, F. C. Proceedings of the First Clinical ACTH Conference, p. 335. Philadelphia, 1950. Blakiston Co.
23. Pearson, O. H. and Eliel, L. P. Experimental studies with ACTH and cortisone in patients with neoplastic disease. In: Recent Progress in Hormone Research, p. 373. New York, 1951. Academic Press.
24. Pearson, O. H., West, C. D., Hollander, V. P. and Escher, G. C. Alterations in calcium metabolism in patients with osteolytic tumors. *J. Clin. Endocrinol.*, 12: 926, 1952.
25. Laszlo, D., Schulman, C. A., Bellin, J., Gottesman, E. D. and Schilling, A. Mineral and protein metabolism in osteolytic metastases. *J. A. M. A.*, 148: 1027, 1952.
26. Bauer, W. and Aub, J. Inorganic salt metabolism. *J. Am. Dietet. A.*, 3: 106, 1927.
27. Albright, F. and Reifenstein, E. C., Jr. The Parathyroid Glands and Metabolic Bone Disease, p. 75. Baltimore, 1948. Williams & Wilkins Co.
28. Heaney, R. P. and Eliel, L. P. Metabolic and cytochemical changes produced by 6-mercaptopurine in human acute leukemia. *Cancer*, 9: 252, 1956.
29. Barker, S. B., Humphrey, M. J. and Soley, M. H. The clinical determination of protein-bound iodine. *J. Clin. Investigation*, 30: 55, 1951.
30. Werner, S. C. The Thyroid, p. 206. New York, 1955. Paul B. Hoeber, Inc.

Thrombohemolytic Thrombocytopenic Purpura*

Case Report and Review of Literature

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ANY disease whose etiology has for thirty-two years been obscure warrants continuing appraisal. The clinical syndrome in question was described first by Moschcowitz [*1*] in 1925. Singer and co-workers [*2*] suggested the name "thrombotic thrombocytopenic purpura." However, since this designation made no reference to the ever present hemolytic anemia, a modification of this name to "thrombohemolytic thrombocytopenic purpura" was proposed by Adelson and co-workers [*3*]. Only forty-six cases of this syndrome have been described in the literature up to 1954 [*3*]. As the disease has become better known, the diagnosis has been made more often before death. The present report describes a case which was correctly diagnosed antemortem.

CASE REPORT

A nineteen year old student nurse was admitted to St. Vincent's Hospital on December 28, 1956, with the chief complaint of weakness and jaundice. She had been in excellent health until two months prior to admission, at which time a lingering "head cold" had developed for which, two weeks later, she received an injection of procaine penicillin. She was then seen by a dentist for gum lesions which were diagnosed tentatively as "trench mouth." Treatment consisted of local gentian violet applications and hydrogen peroxide mouth wash. One month before admission she noted onset of easy fatigability and seemed to require much more rest than usual. Moderately severe frontal headaches developed, for which she took only aspirin and bufferin.® Jaundice was first detected one day before admission, although the patient had noted darker than normal urine for several days, without noticeable change in stool color. On the day of admission she was too weak to stand without assistance, and several large clots of blood were passed vaginally. Her last menstrual period had occurred ten days previously. A few hours before admission she vomited a large amount of yellowish material which contained numerous large blood streaks.

The only drugs taken during the previous three months had been the single injection of procaine penicillin and the aspirin and bufferin.

On admission, the temperature was 101°F.; pulse, 100; respirations, 18; and blood pressure, 110/60 mm. Hg. The patient appeared to be a lethargic, well developed, obese, young woman, who could be easily aroused. Her color was a combination of generalized pallor and jaundice. Several small ecchymoses were present on her arms and legs. The lymph nodes were not palpably enlarged. The liver and spleen were both palpable 1 fingerbreadth below the costal margins and were slightly tender. There was a moderate amount of vaginal bleeding, mostly in the form of clots. Funduscopic examination revealed several fresh retinal hemorrhages bilaterally.

On admission, the red cell count was 1.1 million per cu. mm.; hematocrit, 18 per cent; and hemoglobin, 6 gm. per cent. Red blood cells showed marked anisocytosis, poikilocytosis and hypochromia. The total white cell count was 11,450 per cu. mm., with pronounced shift to the left. The differential count revealed 39 stab forms, 5 juvenile forms, 23 segmented forms, 30 lymphocytes and 3 monocytes. Numerous normoblasts were seen (23 per 100 white blood cells). Bleeding time was one minute and twenty seconds, and clotting time three minutes and fifteen seconds. The direct van den Bergh bilirubin was 0.5 and indirect was 5.6 mg. per cent. The prothrombin time was fourteen seconds with a control at thirteen; blood urea nitrogen, 23 mg. per cent; cephalin flocculation test, 3 plus in forty-eight hours; thymol turbidity test, 6 units; and alkaline phosphatase, 2 units. The urine gave a positive test for free hemoglobin, with a negative urobilinogen test. A serological test for syphilis was negative as was the direct Coombs' test, while the indirect Coombs' test was negative for saline antibodies but positive for blocking antibodies. A platelet

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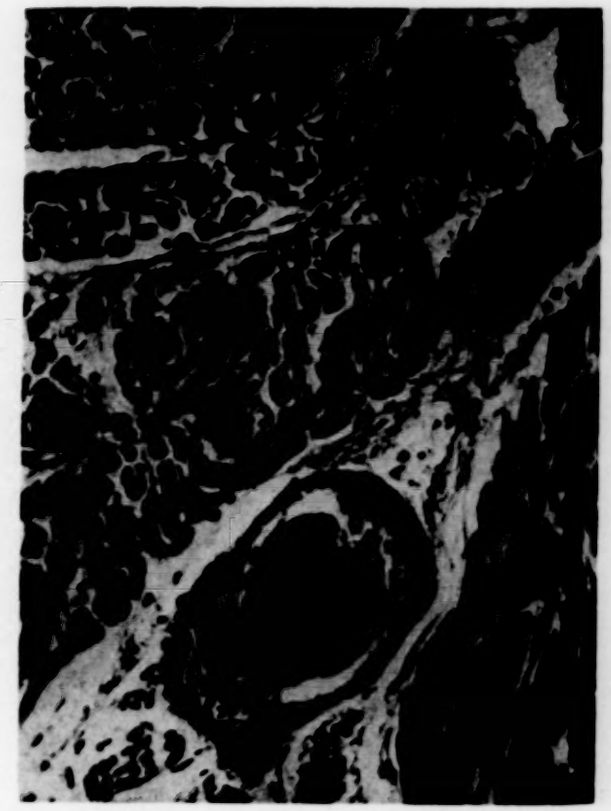

FIG. 1. Section of heart muscle showing occlusion of arterioles with hyaline thrombi.

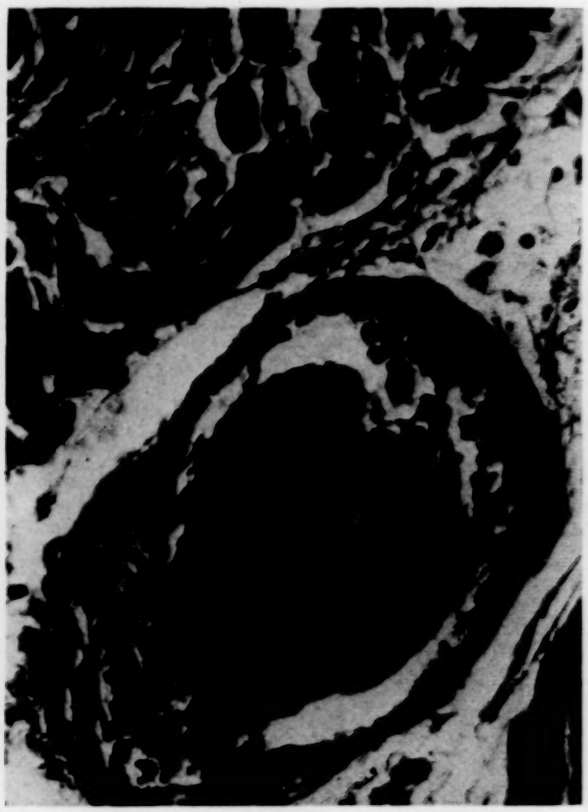

FIG. 2. Arteriole in myocardium showing swelling and proliferation of endothelium as well as obliteration of lumen by hyaline thrombus.

count was 50,000 and there were 15.2 per cent reticulocytes.

The patient's course was progressively downhill despite intensive treatment with multiple whole blood and packed red cell transfusions, ACTH, prednisone, intravenous hydrocortisone and antibiotics. Several hours after admission she lapsed into a semi-comatose state which then alternated with periods of mild delirium and uncooperativeness. There was a suggestion of nuchal rigidity and the reflexes were all hypoactive. Her temperature rose to over 104°F. on the second hospital day and she complained of dizziness and nausea during brief lucid intervals. An increasing number of ecchymoses and petechiae were noted over her abdomen and thigh areas. She died less than forty-eight hours after admission, just before a bone marrow aspiration was to be performed. An antemortem diagnosis of thrombohemolytic thrombocytopenic purpura was made.

The gross general findings at autopsy were those of a young, obese, well developed nineteen year old female measuring 63 inches in length. The body was jaundiced, and ecchymoses and petechiae of arms, legs and abdomen were present. The peritoneal, pericardial and pleural cavities each contained a moderate amount of dark brown-red turbid fluid.

Examination of the heart revealed numerous petechial hemorrhages on the epicardial surfaces. The heart weighed 370 gm. No abnormalities of the valves or chambers were found. The myocardium of the right atrium and pulmonary conus exhibited prominent subendocardial ecchymosis and petechial hemorrhages. The microscopic sections of the myocardium presented a widespread process involving small arterioles and capillaries, in which were noted swelling and proliferation of the endothelium, and obliteration of the lumen of many of these vessels by eosinophilic, hyaline, slightly granular thrombi. (Figs. 1, 2.) The veins were not involved and the process was limited to precapillary arterioles and capillaries. These thrombi were in different stages of organization, containing elongated cells with spindle-shaped nuclei which appeared to arise from the lining of the involved vessels. Extensive foci of interstitial hemorrhage and diffuse edema of the interstitial spaces were noted. The endocardium showed no histopathologic changes.

The liver weighed 1,990 gm. and presented a smooth, glistening capsule with many petechial hemorrhages. Sections showed a normal lobular architecture. The portal spaces revealed marked lymphocytic infiltration. Numerous branches of the hepatic arteries and arterioles showed thrombotic processes similar to those described in the heart. (Fig. 3.) Here also, as in the heart, the endothelial changes were prominent even in the non-thrombosed vessels.

FIG. 3. Section of liver with an arteriole occluded by a hyaline thrombus.

The hepatic lobules exhibited prominent central congestion associated with focal areas of central necrosis. Many giant cells were seen within the hepatic sinusoids.

The spleen was considerably enlarged and weighed 475 gm. Sections of this organ revealed considerable engorgement of the sinusoids of the red pulp. The arteries were surrounded by abundant lymphoid tissue with prominent germinal centers which contained aggregates of numerous, large mononuclear cells.

The right adrenal gland weighed 6.5 gm. and the left 7 gm. Sections showed prominent degrees of thrombosis of the vessels as seen in the heart and other organs. This process was limited almost entirely to the zona glomerulosa. Sections of the pancreas also revealed numerous hyaline thrombi of capillaries and precapillary arterioles. Sections of the gastrointestinal tract disclosed no histopathologic changes other than engorgement of the blood vessels. Lung sections revealed only varying degrees of engorgement of the pulmonary vasculature, distention of alveoli with protein rich fluid, and engorged interalveolar septums containing numerous giant cells.

Microscopic examination of the kidney showed thrombosis of an occasional interlobular arteriole with an eosinophilic hyaline substance. The glomeruli appeared hypercellular and many of the glomerular capillaries contained hyaline thrombi.

The lymph nodes showed well preserved architecture, with a considerable degree of blood vessel engorgement. Throughout the lymphoid tissue and sinuses, many giant cells were visible and hyaline thrombosis of arterioles and capillaries was prominent, as were proliferative changes of the blood vessel walls. The bone marrow was extremely hypercellular, with almost complete absence of fatty vacuoles. Hyperplasia of all bone marrow elements contributed to the hypercellularity. Megakaryocytes were particularly prominent and numerous, 16 per high power field being seen in some areas. Hyaline thrombosis of capillaries was occasionally found. Permission to examine the brain was not granted.

COMMENTS

The pathognomonic histologic findings in thrombohemolytic thrombocytopenic purpura consist of a homogeneous eosinophilic material partially or completely occluding arterioles, capillaries and, occasionally, venules throughout the body [*3*]. Moschcowitz [*1*] suggested originally that this material was thrombus formed by agglutinated red blood cells. Baehr and coworkers [*4*] postulated that the plugs were composed of platelets trapped in the small blood vessels, thereby producing a fall in the level of circulating platelets. There are at present no specific stains available for platelet differentiation. It was first suggested by Altschule [*5*] that the platelet thrombosis might be secondary to the primary defect of endothelial cell proliferation. Since then several workers have emphasized the importance of the associated changes in the endothelium of the occluded vessels. The intraluminal masses, which have a bimorphous hyaline and granular appearance, were considered by Gore [*13*] to represent an extruded intramural material with superimposed platelet thrombi.

In the presently described case it appeared that the lesions were due to a combination of findings expressed by both schools of thought, namely, swelling and proliferation of the endothelium as well as obliteration of the lumen of these vessels by eosinophilic hyaline thrombi.

The distribution of lesions and their severity varies, but any organ may be involved. The most extensively affected organs, as determined by Adelson and co-workers [*3*], are heart, brain, adrenal cortex, lymph nodes, kidney, liver and spleen.

It has been stated that three of every five cases of this condition occur in females, and that two-thirds of those affected are between the ages of ten and forty [*3*]. The patient reported in the present paper was a nineteen year old female whose past history was completely negative for any allergy. However, several cases have been reported with histories of allergic tendencies, including penicillin sensitivity [*10*], hypertrophic rhinitis [*14*] and sulfa sensitivity with urticaria. Iodine allergy was suggested in the etiology of one case [*16*]. The actual cause of the disease process is, however, still unknown, but it can best be summarized as a hypersensitivity state involving red blood cells, platelets, megakaryocytes and vessel walls [*3*]. Various prodromata [*4,15,17*] have been recorded in this condition, including ten cases of upper respiratory infections just prior to the onset of symptoms, two cases of urticaria, and one of dermatitis. Also, administration of tetanus antitoxin [*18*] and smallpox vaccination [*19*] were reported just preceding onset of symptoms in two cases. In our case the only prodrome was a mild, lingering "head-cold," and no medications had been taken other than aspirin, bufferin and a single penicillin injection. All these medications had also been taken in the past without any untoward reactions.

Of the forty-nine patients collected from the literature and reviewed by Adelson [*3*], forty-four had diffuse and multiple neurological manifestations, consisting of motor paralyses, seizures or disturbances in the state of consciousness. Evidence of peripheral nerve and spinal cord damage has been recorded [*20*]. Slight nuchal rigidity, hypoactive reflexes and lethargy which changed to mild delerium were all manifested by our patient.

Eight patients were treated with steroids but the outcome was unsuccessful in all of them. Likewise in our case no improvement resulted from the use of intravenous hydrocortisone and oral prednisone. Meacham and co-workers [*21*] have reported temporary improvement in one case following splenectomy.

Hemorrhagic manifestations have been described in most cases, and in our case skin petechiae and ecchymoses, hematuria, vaginal bleeding and retinal hemorrhages all made their appearance.

Among the reported cases, the red cell count has varied from 1 million [*21*] to over 5 million [*22*], and hemoglobin values have ranged from 18 per cent [*4*] to 78 per cent [*22*]. In the present case the red cell count was 1.1 million per cu. mm. with a hemoglobin of 6 gm. per cent. Also, numerous normoblasts (23 per 100 white blood cells) were noted, as in many of the previously described cases. Most of the reported patients have revealed an elevated reticulocyte count [*23*] which, in our case, was 15.2 per cent. The differential white blood cell count in our case revealed a pronounced shift to the left with 5 juvenile and 39 stab forms. In thirty-eight of forty-one reported cases [*3*], similar types of differential counts were recorded. In this same group of cases platelet counts ranged from 10,000 to 100,000. In our case the platelet count was 50,000. The elevated serum bilirubin in the case is indicative of the hemolytic process.

This clinical syndrome is known to be rapidly fatal despite all therapy. The duration in the present case, from the onset of the prodromal respiratory symptoms to the final stage of delirium and death, was approximately two months. Recently, however, Báez-Villaseñor [*24*] recorded a case of a forty-three year old woman with this disease who had had episodes of purpura for eight years. It was pointed out by Singer [*25*] that in only two cases in the literature which he reviewed was there a chronic course.

Another interesting fact, worthy of pointing out, is that the Coombs' test is ordinarily negative in this condition. This test, however, was positive in a recently reported case of thrombotic thrombocytopenic purpura [*24*]. A positive Coomb's test indicates that the hemolysis is due to extra-erythrocytic factors. In our case, although the direct Coombs' test was negative, the indirect Coombs' was positive for blocking antibodies. Nussbaum and Dameshek [*26*] have just published a case in which there was a transient disturbance of the type of thrombohemolytic thrombocytopenic purpura during the course of meningococcemia which may have been mediated through an indirect or immunologic mechanism. These authors emphasize that further investigation of the possible etiological role of infection is warranted. In fact, recently, by injecting meningococcal endotoxin into rabbits Brunson and co-workers [*27,28*] have noted lesions which are similar to those of thrombohemolytic thrombocytopenic purpura.

Bone marrow examination in the present case was performed postmortem and revealed the picture typical of this condition, namely, hypercellularity with an increase in megakaryocytes

without much platelet formation. The antemortem diagnosis of thrombohemolytic thrombocytopenic purpura has usually been established by isolation of vessels with characteristic lesions in bone marrow specimens or by splenectomy, but recently it has been found that biopsy of random lymph nodes during life may yield tissue of diagnostic histology [*29*].

SUMMARY

A case is presented of a nineteen year old female in whom an antemortem diagnosis of thrombohemolytic thrombocytopenic purpura was made and substantiated at necropsy. The clinical picture, which combined hemolytic anemia, thrombocytopenic purpura and cerebral symptoms, suggested the correct diagnosis.

The organs most affected were the heart, liver, kidneys, spleen and adrenals. Veins were not involved, and the process was limited to the precapillary arterioles and capillaries. The lesions consisted of a combination of obliteration of the vessel lumen by eosinophilic, hyaline thrombi and proliferation of the endothelium itself.

The absence of an etiology and the inevitable downhill course despite steroid therapy conforms with previous results noted in reviewing the literature.

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REFERENCES

1. Moschcowitz, E. Acute febrile pleiochromic anemia with hyaline thrombosis of terminal arterioles and capillaries. *Arch. Int. Med.*, 36: 89–93, 1925.
2. Singer, K., Bornstein, F. P. and Wile, S. A. Thrombotic thrombocytopenic purpura: hemorrhagic diatheses with generalized platelet thromboses. *Blood*, 2: 542–554, 1947.
3. Adelson, E., Heitzman, E. J. and Fennessey, J. F. Thrombohemolytic thrombocytopenic purpura. *Arch. Int. Med.*, 94: 42–60, 1954.
4. Baehr, G., Klemperer, P. and Schifrin, A. An acute febrile anemia and thrombocytopenic purpura with diffuse platelet thromboses of capillaries and arterioles. *Tr. A. Am. Physicians*, 51: 43–58, 1936.
5. Altschule, M. D. A rare type of acute thrombocytopenic purpura: widespread formation of platelet thrombi in capillaries. *New England J. Med.*, 227: 447–479, 1942.
6. Trobaugh, F. E., Markowitz, M., Davidson, C. and Crowley, W. An acute febrile illness characterized by thrombocytopenic purpura, hemolytic anemia, and generalized platelet thrombi. *Arch. Path.*, 41: 327–334, 1946.
7. Carter, J. R. Generalized capillary and arteriolar platelet thrombosis. *Am. J. M. Sc.*, 213: 585–593, 1947.
8. Engel, G. L., Scheinker, I. M. and Humphrey, D. C. Acute febrile anemia and thrombocytopenia. with vasothromboses. *Ann. Int. Med.*, 26: 919–933, 1947.
9. Muirhead, E. E., Grass, G. and Hill, J. M. Diffuse platelet thromboses with thrombocytopenia and hemolytic anemia (thrombotic thrombocytopenic purpura). *Am. J. Clin. Path.*, 18: 523–532, 1948.
10. Gendel, B. R., Young, J. M. and Kraus, A. P. Thrombotic thrombocytopenic purpura. *Am. J. Med.*, 13: 3–11, 1952.
11. Cooper, T., Stickney, J. M., Pease, G. L. and Bennett, W. A. Thrombotic thrombocytopenic purpura. *Am. J. Med.*, 13: 374–383, 1952.
12. Orbison, J. L. Morphology of thrombotic thrombocytopenic purpura with demonstration of aneurysms. *Am. J. Path.*, 28: 129–143, 1952.
13. Gore, I. Disseminated arteriolar and capillary platelet thrombosis: a morphologic study of its histogenesis. *Am. J. Path.*, 26: 155–175, 1950.
14. Beigelman, P. M. Variants of the platelet thrombosis syndrome and their relationship to disseminated lupus. *Arch. Path.*, 51: 213–223, 1951.
15. Fitzgerald, P. I. Auerbach, O. and Frame, E. Thrombotic acroangiothrombosis (platelet thromboses of the capillaries, arterioles and venules). *Blood*, 2: 519–541, 1947.
16. Ehrich, W. E. and Seifter, J. Thrombotic thrombopenic purpura caused by iodine: report of a case. *Arch. Path.*, 47: 446–449, 1949.
17. Bernheim, A. I. Widespread capillary and arteriolar platelet thrombi. *J. Mt. Sinai Hosp.*, 10: 287–291, 1943.
18. Barondess, J. A. Thrombotic thrombocytopenic purpura. *Am. J. Med.*, 13: 294–310, 1952.
19. Green, M. A. and Rosenthal, S. Generalized blood platelet thrombosis: report of 3 cases with necropsy findings. *J. Mt. Sinai Hosp.*, 16: 110–120, 1949.
20. Adams, R. D. Cammermeyer, J. and Fitzgerald, P. J. The neuropathological aspects of thrombocytic acroangiothrombosis. *J. Neurol., Neurosurg. & Psychiat.*, 11: 27–43, 1948.
21. Meacham, G. C., Orbeson, J. L., Heinle, R. W., Stelle, H. J. and Schaefer, J. A. Thrombotic thrombocytopenic purpura: a disseminated disease of arterioles. *Blood*, 6: 706–719, 1951.
22. Brown, E. E. and Norman, J. W. Multiple platelet thrombi (clinicopathological conference). *New York J. Med.*, 46: 2167, 1946.
23. Gitlow, S. and Goldmark, C. Generalized capillary and arteriolar thrombosis: report of two cases with a discussion of the literature. *Ann. Int. Med.*, 13: 1046–1067, 1939.
24. Báez-Villaseñor, J. and Ambrosius, K. Thrombotic thrombocytopenic purpura: report of a case with some unusual characteristics. *Ann. Int. Med.*, 46: 378–386, 1957.
25. Singer, K. Thrombotic thrombocytopenic purpura. *Advance Int. Med.*, 6: 195, 1954.
26. Nussbaum, M. and Dameshek, W. Transient hemolytic and thrombocytopenic episode (? acute transient thrombohemolytic thrombocytopenic

purpura) with probable meningococcemia. *New England J. Med.*, 256: 448–450, 1957.

27. Brunson, J. G., Thomas, L. and Gamble, C. N. Morphologic changes in rabbits following intravenous administration of meningococcal toxin. II. Two appropriately spaced injections: role of fibrinoid in generalized Schwartzman reaction. *Am. J. Path.*, 31: 655–667, 1955.
28. Brunson, J. G., Davis, R. L. and Thomas, L. Morphologic changes in rabbits following intravenous administration of meningococcal toxin. III. Effects produced by endotoxin in association with certain high molecular weight acidic polymers. *Am. J. Path.*, 31: 669–685, 1955.
29. Morey, D. A. J., White, J. B. and Daily, W. M. Thrombotic thrombocytopenic purpura diagnosed by random lymph node biopsy. *Arch. Int. Med.*, 98: 821–823, 1956.

Secondary Amyloidosis and Hepatic Failure in Hodgkin's Disease*

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PROGRESSIVE jaundice culminating in hepatic failure was observed during the last month of illness in a young man with extensive Hodgkin's disease whose liver was massively infiltrated with amyloid. Deposition of amyloid in the liver has been reported in subjects who have Hodgkin's disease, particularly following therapy with nitrogen mustard. Jaundice has been observed occasionally in patients with secondary amyloidosis, but hepatic failure apparently has not been described. The observation of hepatic failure apparently due to amyloidosis in a subject with Hodgkin's disease prompted this brief report.

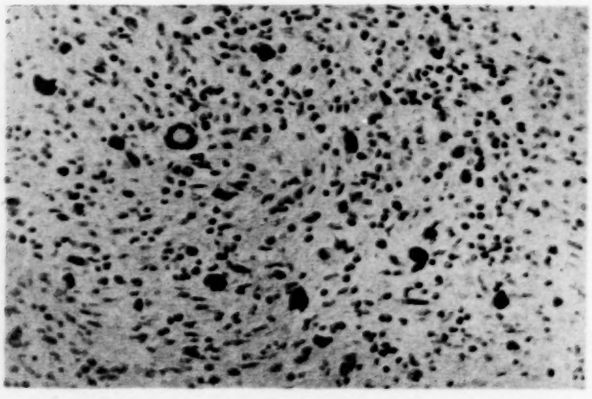

FIG. 1. A photomicrograph of a lymph node completely replaced by Hodgkin's disease. Note the Reed-Sternberg giant cells and reticulum cells in the fibrous stroma. Original magnification × 440, hematoxylin and eosin.

CASE REPORT

A thirty-three year old white heating plant engineer entered the Salt Lake Veterans Administration Hospital on February 28, 1955, complaining of progressive swelling of his neck for three months. One month before this admission he had consulted a physician because of a sore throat and fever which were accompanied by a swollen neck. Treatment consisted of bedrest and penicillin. The throat soreness gradually disappeared but he was admitted to this hospital because of persisting cervical node enlargement and fever. He had also noted a weight loss of 20 pounds, mild anorexia, listlessness and weakness during the two months which preceded admission. The past history was not related to his illness.

Physical examination revealed a pale young man with a thick neck. The temperature was 102°F; the pulse rate 100; blood pressure 110/76 mm. Hg; and respiratory rate 16. Enlarged non-tender lymph nodes were present in both anterior cervical chains, both axillae and the left inguinal area. A mass of matted lymph nodes was present in the right inguinal area. The spleen and liver were not palpable.

The volume of packed red blood cells was 35 ml. per 100 ml. and the white blood cell count was 8,700 per cu. mm. with 81 per cent polymorphonuclears, 5 per cent lymphocytes and 13 per cent monocytes. The red blood cells appeared normochromic and normocytic on a stained smear. Examination of the urine revealed nothing remarkable. The serologic test for syphilis and the first strength tuberculin test were negative. The other laboratory findings are shown in Table 1. A pathologic diagnosis of Hodgkin's disease was made by biopsy of a large supraclavicular lymph node. (Fig. 1.)

The temperature gradually returned to normal without treatment. Therapy with 38 mg. (0.6 mg. per kg. of body weight) of methyl bis-(beta-chlorethyl)-amine hydrochloride (nitrogen mustard) was given. This was followed by subjective improvement of the patient and a reduction in the size of the lymph nodes.

During the next twelve months the patient had six admissions to the hospital for exacerbations of his disease and received a total of 2,700 roentgens of radiation to both cervical areas and the left axilla and an additional 160 mg. of nitrogen mustard. The clinical and laboratory findings on these admissions are summarized in Table 1.

Because of fever, a hacking cough, exertional dyspnea, a weight loss of sixteen pounds and an increase in the size of his cervical and axillary nodes the patient was admitted for the eighth time on June 6,

* From the Veterans Administration Hospital and the Department of Pathology of the University of Utah College of Medicine, Salt Lake City, Utah.

TABLE I

CLINICAL AND LABORATORY FINDINGS

Data	First Admission (2/28/55–3/16/55)		Second Admission (8/15/55)	Third Admission (10/25/55–11/7/55)		Fourth Admission (1/4/56)	Fifth Admission (2/6/56)	Sixth Admission (3/22/56)	Seventh Admission (5/28/56)	Eighth Admission (6/6/56)	Ninth Admission (8/24/56)	
Clinical data:												
Weight	145	.....	170	178	.....	177	178	.....	.....	157	147	
Temperature, °F.	102.2	98.6	98.6	100	.....	104	99.6	.....	.....	102	101.4	
Nodes	++	+	+	++	++	++	++	++	+++	+++	++++	
Liver } below costal margin	.....	.....	.....	.....	.....	.....	.....	.....	.....	4 cm.	8 cm.	
Spleen } below costal margin	.....	.....	.....	.....	.....	.....	.....	.....	.....	4 cm.	4 cm.	
Therapy:												
Nitrogen mustard	38 mg.		46 mg.	49 mg.	.....	.....	28 mg.	.....	.....	28 mg.	13 mg.	
X-ray therapy	.....	.....	.....	.....	.....	1200 R	.....	1500 R	.....	.....	.....	
Laboratory data:												
Volume of packed red blood cells, ml./100 ml.	35	.....	42	42	34	39	39	35	31	27.5, 21*	18-36†	
White blood cells/cu. min.	8,700	4,700	8,400	7,600	2,075	7,600	.....	6,100	.....	4,750‡	5,000	
Differential:												
Polymorphonuclears	81	.....	79	68	64	72	.....	73	.....	95	98	
Lymphocytes	5	.....	10	25	32	22	.....	19	.....	3	1	
Monocytes	13	.....	6	4	2	5	.....	3	.....	2	1	
Eosinophils	.....	.....	2	.....	.....	1	.....	4	.....	.....	.....	
Basophils	1	.....	3	3	2	.....	.....	1	.....	.....	.....	
Urine:												
Specific gravity	1.023	.....	.....	.....	.....	.....	.....	.....	.....	.....	1.025	1.012
Albuminuria	0	.....	.....	.....	.....	.....	.....	.....	.....	0	0	0
Bile	0	.....	.....	.....	.....	.....	.....	.....	.....	0	2†	4†
Chemistries:												
BSP 45-minute retention	.....	.....	.....	.....	.....	3.5%	.....	.....	.....	20%	.....	
Bilirubinmg. /100 ml., d/t	.....	.....	.....	.....	.....	0.2/0.9	.....	.....	.....	0.6/1.8	4.9/8.3	8.8/13.8
Alkaline phosphatase, mg./100 ml.	.....	.....	.....	.....	.....	3.4	.....	.....	.....	16,12,7	4	.....
Serum proteins, total/albumin gm./100 ml.	.....	7.0/4.3	.....	8.0/4.7	.....	7.6/4.6	.....	.....	.....	6.6/3.5	3.1/1.7	2.7/1.5
Prothrombin time	.....	.....	.....	.....	.....	100%	.....	.....	.....	100%	16%	.....
Cr^{51} erythrocyte half-life	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	11 days (normal 26 days)	

* Transfused with 4 L. whole blood.
† Transfused with 3 L. whole blood.
‡ White blood cell count fell to 185/cu. mm.

1956. On admission he had a temperature of 102°F and enlargement of superficial lymph nodes. The liver edge and the tip of the spleen were both palpable.

The volume of packed red blood cells was 27.5 ml. per 100 ml., the white blood cell count was 4,750 per cu. mm. with 95 per cent polymorphonuclears, and 5 per cent lymphocytes. The bromsulphalein® retention was 20 per cent in forty-five minutes, the serum bilirubin concentration was 0.6 mg. direct and 1.8 mg. total per 100 ml. The serum alkaline phosphatase level was 16 and later 12 Shinowara-Jones-Reinhart units per 100 ml. The total serum protein level was 6.6 gm. with 3.5 gm. of albumin per 100 ml. (Table I.)

Twenty-six mg. (0.4 mg. per kg.) of nitrogen mustard were administered. Six days after therapy the white blood count had dropped to 185 per cu. mm. and the volume of packed red blood cells was 21 ml. per 100 ml.

There was no evidence of blood loss to explain this persistent anemia. The osmotic fragility of the red blood cells was normal. The tests for acid, cold and warm hemolysins and cold and warm agglutinins were negative. The Donath-Landsteiner test was negative. The direct and indirect Coombs' tests were also negative. The patient was given 4 L. of whole blood which raised the volume of packed red blood cells to 36 ml. per 100 ml. The half-life of the patient's Cr^{51}-tagged erythrocytes was eleven days compared to an erythrocyte half-life of twenty-six days for a normal subject. Therapy with 50 mg. of prednisolone per day for two weeks had no effect on the rate of blood destruction.

Because of weakness and exertional dyspnea, the patient was readmitted on August 24, 1956. He was pale, jaundiced and appeared acutely ill. There was pitting edema of the ankles. Both the liver and spleen showed a striking increase in size and were palpable 8 cm. and 4 cm., respectively, below the costal margin.

The volume of packed red blood cells was 18 ml. per 100 ml. and the white blood cell count was 5,000 per cu. mm. with 96 per cent polymorphonuclear cells. The test for bile in the urine was strongly positive. The serum bilirubin concentration was 4.9 mg. direct and 8.3 mg. total per 100 ml. The serum alkaline phosphatase level was 4.0 Shinowara-Jones-Reinhart

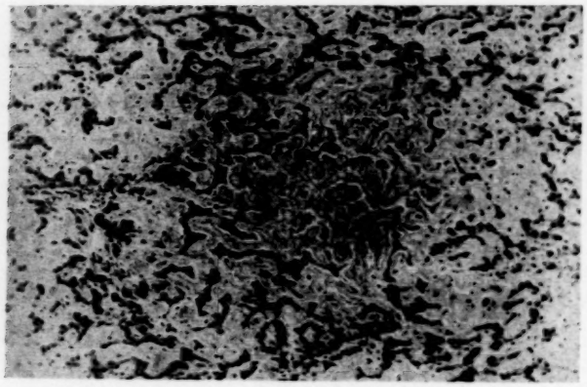

FIG. 2. A photomicrograph of the liver showing the marked amyloid deposition. Original magnification × 200, hematoxylin and eosin.

FIG. 3. A photomicrograph of the liver showing one of the periportal nodules of Hodgkin's disease and deposits of amyloid. Original magnification × 200, hematoxylin and eosin.

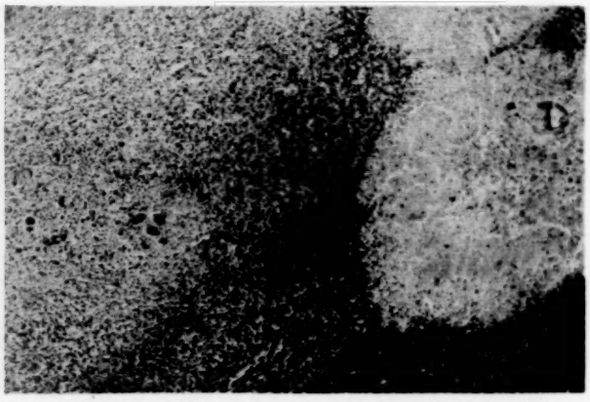

FIG. 4. A photomicrograph of the spleen showing Hodgkin's disease. Note the giant cells and the amyloid deposits around an arteriole. Original magnification × 100, hematoxylin and eosin.

units per 100 ml. and the total serum protein level was 4.0 gm. with 2.4 gm. of albumin per 100 ml.

Because hepatic involvement from Hodgkin's disease was thought to be the best explanation of this jaundice, on September 5, 1956, 13 mg. of nitrogen mustard were administered. No benefit was apparent during the next five days. The blood pressure had fallen to 90/60 mm. Hg. The patient became increasingly edematous and oliguric. The serum albumin level dropped to 1.5 gm. per 100 ml. and the serum sodium concentration fell to 119 mEq. per L. He passed several tarry stools and became more jaundiced. The plasma prothrombin time fell to 16 per cent of normal but the levels of alkaline phosphatase and the thymol turbidity did not rise. A flapping tremor developed and the patient became extremely drowsy. Bile, granular bile casts and albumin were present in the urine. Gastric suction was productive of bright red blood and melena continued.

On September 8, the patient had an episode of pulmonary edema followed by coma, marked hypotension and anuria. Despite supportive measures which included intravenous arterenol he died the following day.

At autopsy, the body was markedly icteric and showed generalized pitting edema. Tumor nodules were scattered through the lungs and pleura and particularly in the peribronchial lymph nodes. The heart was within normal limits.

The peritoneal cavity contained 3,000 ml. of straw-colored fluid. The liver weighed 2,250 gm. and a few small tumor nodules were present on the capsule. The extrahepatic biliary system showed no obstruction and bile was easily expressed into the duodenum. Microscopic examination of the liver (Fig. 2) revealed extensive deposits of homogeneous eosinophilic material in sinusoids and around hepatic cells. The material was stained pink by hematoxylin-eosin and by Congo red and gave a typical staining reaction with crystal violet. It filled most of the sinusoids of the liver and surrounded portal triads. The remaining hepatic cells appeared vacuolated and compressed. Amyloidosis was the predominant finding although there were several small periportal nodules of Hodgkin's disease. (Fig. 3.)

The spleen was nodular and weighed 640 gm. Many pale tan nodules appeared necrotic and the intervening parenchyma was reddish brown without distinct follicles. The splenic arterioles were surrounded by homogeneous pale eosinophilic material. (Fig. 4.) The kidneys weighed 200 and 180 gm. and were soft, swollen and the sectional surfaces showed a radial arrangement of green pigment. The epithelial cells of the proximal convoluted tubules and the loops of Henle were deeply bile stained. The cellular outlines were indistinct and fused with one another. Homogeneous dense yellow casts filled many collecting tubules. A few glomeruli contained small deposits of amyloid. Many lymph nodes were enlarged by yellow necrotic tumor nodules up to 3 cm. in diameter and on microscopic examination rims of intact tumor surrounded foci of pale eosinophilic material. Some areas in the bone marrow were pale and dry but other areas

were filled with yellow necrotic tumor. Examination of the sections from the ribs and vertebral bodies showed foci of viable and necrotic tumor and only occasional hemopoietic cells.

COMMENTS

Various authors [*1–3*] have reported jaundice in from 3 to 58 per cent of patients with Hodgkin's disease. Jaundice was observed in 68 per cent (fifteen of twenty-two) of such patients who died at this hospital [*4*]. The mechanism producing jaundice is frequently obscure and may include a combination of prehepatic, posthepatic and hepatocellular factors. The cause of prehepatic jaundice is usually hemolytic anemia. Secondary hemolytic anemia has been reported in Hodgkin's disease [*5,6*] but it only occasionally causes slight jaundice.

Encroachment on the biliary system by invasion or by compression by involved perihilar nodes is commonly postulated to explain findings which suggest posthepatic jaundice in Hodgkin's disease. Invasion of the extrahepatic biliary tract has been described in several reports [*4,7*] but jaundice was present only in the case reported by Middleton [*8*] and in one patient observed at this hospital. In both of these there was compression of the hepatic ducts in the hilum of the liver by enlarged lymph nodes which contained Hodgkin's disease. Beatty [*1*] described two patients with apparent extrahepatic biliary obstruction due to Hodgkin's disease in the hilar lymph nodes but he found no microscopic evidence of biliary obstruction in the liver.

The jaundice in Hodgkin's disease is usually hepatocellular in origin but may be due chiefly to intrahepatic biliary tract obstruction as a consequence of infiltration of the liver. However, in some cases jaundice can be attributed to a coexisting but unrelated disease in the liver particularly viral hepatitis [*4,9*]. Although the clinical recognition of viral hepatitis in a patient with Hodgkin's disease is often difficult the pathologic differences are unquestionable. Because 50 to 95 per cent of patients with Hodgkin's disease have evidence of hepatic involvement by tumor, it is somewhat surprising that the incidence of jaundice is not higher than that usually reported. The histologic pattern is that of a periportal infiltrate which causes compression and necrosis of hepatic cells. Of forty patients reported by Beatty [*1*], twenty-three were jaundiced and periportal infiltration by tumor was consistently present. Similar hepatic infiltrates were present in fifteen patients with jaundice from a total of twenty-two patients with Hodgkin's disease autopsied at this hospital [*4*].

Amyloidosis occurring secondary to Hodgkin's disease can involve the liver although renal amyloidosis associated with a nephrotic syndrome has been recognized more often. In a review in 1950 Wallace and associates [*10*] accepted thirty-five cases of amyloidosis secondary to Hodgkin's disease from the world literature. Several cases of amyloid nephrosis occurring in subjects with Hodgkin's disease treated with nitrogen mustard have been reported since 1948 [*11*].

Jaundice and hepatic failure are rarely observed in hepatic amyloidosis although bromsulphalein retention and slightly elevated icteric indices without clinical jaundice are common in patients with amyloidosis secondary to tuberculosis [*12–14*]. One patient with amyloidosis secondary to Hodgkin's disease developed jaundice which appeared to remit before his death. At autopsy massive deposits of amyloid were present in the liver [*1*]. Bannick [*15*] reported a patient with mild jaundice who had amyloidosis secondary to a gastric carcinoma. Spain and Riley [*16*] found involvement of the liver in fifty of seventy-eight patients with secondary amyloidosis but only one showed clinical jaundice. This patient had extensive pulmonary tuberculosis; a large liver developed and an increased serum alkaline phosphatase level and he became clinically jaundiced one week before his death. At autopsy the liver weighed 3,200 gm. and was extensively infiltrated by amyloid. The replacement of this liver by amyloid greatly exceeded the infiltration present in the livers of the other patients in this series. The authors suggested that a rapid deposition of amyloid had produced both a numerical insufficiency of hepatic cells and obstruction of some bile canaliculi resulting in jaundice. In several recent case reports and in animal experiments [*17,18*] the suggestion has been made that the administration of nitrogen mustard accelerates the deposition of amyloid by depressing or destroying actively proliferating mesenchymal cells. In one experiment, mice given sodium caseinate and nitrogen mustard showed amyloidosis earlier and to a more striking degree than controls who received sodium caseinate alone.

In the present case secondary hemolytic anemia may have been present as was suggested by the shortened half-life of erythrocytes and the

rapid disappearance of transfused blood. The absence of a reticulocyte response to this hemolysis may have been caused by the extensive infiltration of the bone marrow by Hodgkin's disease.

There was no obstruction of the extrahepatic biliary tract and no evidence of obstructive jaundice was present in the liver. There were no stigmas of inflammatory or toxic liver disease, and it would be difficult to explain this degree of jaundice by the rare periportal foci of Hodgkin's disease.

A reconstruction of the events in this case suggests that the extensive amyloid deposits in the liver caused a significant decrease in the number of hepatic cells and compression of those remaining, with obstruction of some bile canaliculi. Analogous to the patient reported by Spain and Riley, it is conceivable that these changes occurring over a short period of time could have produced jaundice and hepatic failure. A fairly abrupt deposition is suggested by the rapid increase in the size of the liver with a fulminant loss of hepatic function manifested by progressive jaundice, ascites, hepatic coma and death. A total of 200 mg. of nitrogen mustard was administered to this patient and may have been a factor in accelerating the deposition of amyloid.

SUMMARY

A young man with Hodgkin's disease died of hepatic failure. At autopsy masses of tumor were found in the skin, lymph nodes, lungs, pleura, spleen and bone marrow. The hepatic cells were replaced or compressed by deposits of amyloid and only a few portal triads were infiltrated by Hodgkin's disease. It is postulated that the accumulation of amyloid was associated with extensive Hodgkin's disease and nitrogen mustard therapy. The hepatic failure evidenced by progressive jaundice, ascites, coma and the increase in the size of the liver can be best explained by rapid deposition of massive amounts of amyloid.

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REFERENCES

1. Beatty, E. C., Jr. Jaundice in Hodgkin's disease. *Bull. New York Acad. Med.*, 30: 409–412, 1954.
2. Case records of Massachusetts General Hospital, #35391. *New England J. Med.*, 241: 497–500, 1949.
3. Goldman, L. B. Hodgkin's disease, an analysis of 212 cases. *J. A. M. A.*, 114: 1611–1616, 1940.
4. Kilburn, K. H. Analysis of 22 cases of Hodgkin's disease with jaundice from the Salt Lake Veterans Administration Hospital. (Unpublished.)
5. Brown, R. J. and Meynell, M. J. Haemolytic anemia associated with Hodgkin's disease. *Lancet*, 2: 835–836, 1949.
6. Wasserman, L. R., Stats, D., Schwartz, L. and Fudenberg, H. Symptomatic and hemopathic hemolytic anemia. *Am. J. Med.*, 18: 961–989, 1954.
7. Harrell, G. T. Hodgkin's disease with invasion of the pericardium and gallbladder. *Arch. Path.*, 28: 58–64, 1939.
8. Middleton, W. S. Some clinical caprices of Hodgkin's disease. *Ann. Int. Med.*, 11: 448–468, 1937.
9. Hoster, H. A., Zanes, R. P. and von Haam, E. The association of "viral" hepatitis and Hodgkin's disease. *Cancer Res.*, 9: 473–480, 1949.
10. Wallace, S. L., Feldman, D. J., Berlin, I. and Harris, C. Amyloidosis in Hodgkin's disease. *Am. J. Med.*, 8: 552–557, 1950.
11. Dahlin, D. C. Secondary amyloidosis. *Ann. Int. Med.*, 31: 105–119, 1949.
12. Kruger, A. L. and Gerber, I. E. Bromsulfalein liver function test in pulmonary tuberculosis. *Am. Rev. Tuberc.*, 46: 439–444, 1942.
13. Tiber, A. M., Pearlman, A. W. and Cohn, S. E. Hepatic function in patients with amyloidosis. *Arch. Int. Med.*, 68: 309–324, 1955.
14. Wald, M. H. Clinical studies of secondary amyloidosis in tuberculosis. *Ann. Int. Med.*, 43: 383–395, 1955.
15. Bannick, E. G., Berkman, J. M. and Beaver, D. C. Diffuse amyloidosis. *Arch. Int. Med.*, 51: 978–990, 1933.
16. Spain, D. M. and Riley, R. L. Jaundice in amyloidosis of the liver. *Am. J. Clin. Path.*, 14: 284–288, 1944.
17. Teilum, G. Studies on the pathogenesis of amyloidosis, effect of nitrogen mustard in inducing amyloidosis. *J. Lab. & Clin. Med.*, 43: 367–374, 1954.
18. Spain, D. M. Rapid and extensive development of amyloidosis in association with nitrogen mustard therapy. *Am. J. Clin. Path.*, 26: 52–55, 1956.

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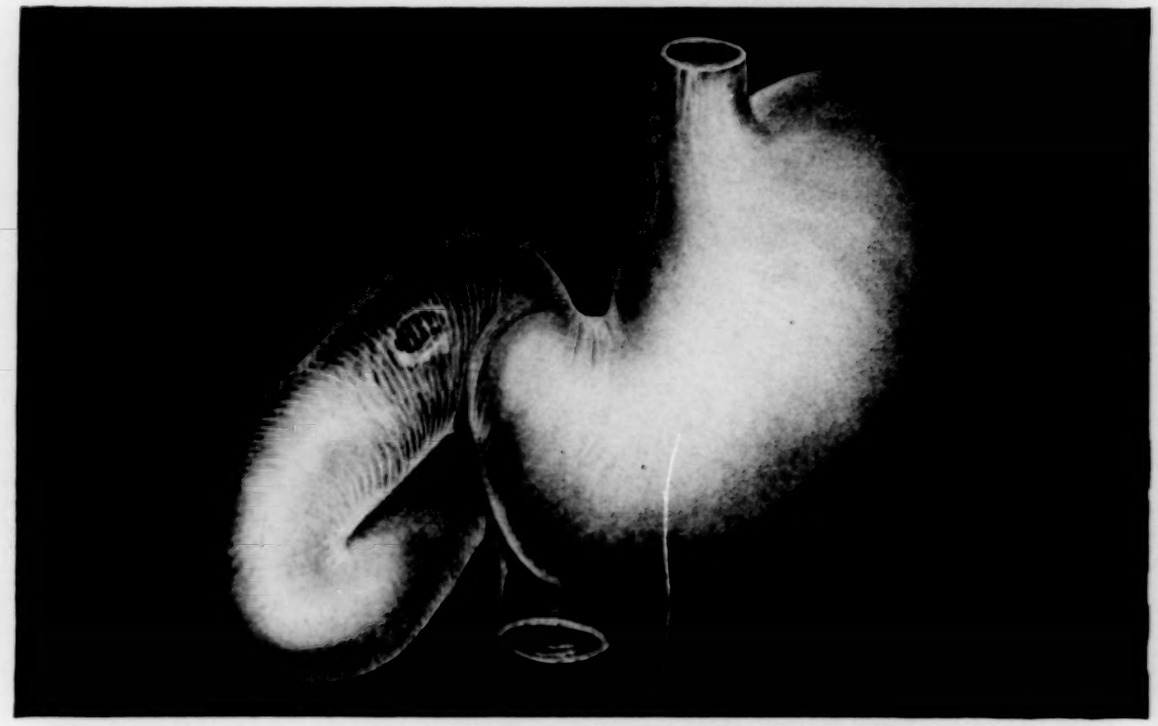

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*Lichstein, J.; Morehouse, M. G., and Osmon, K. L.: Pro-Banthine in the Treatment of Peptic Ulcer. A Clinical Evaluation with Gastric Secretory, Motility and Gastroscopic Studies. Report of 60 Cases, Am. J. M. Sc. *232*:156 (Aug.) 1956.

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1. Cornely, D. A. and Ritter, J. A.: N-acetyl-*p*-aminophenol (Tylenol Elixir) as a Pediatric Antipyretic-Analgesic, J. A. M. A. *160*:1219-1221 (April 7) 1956.

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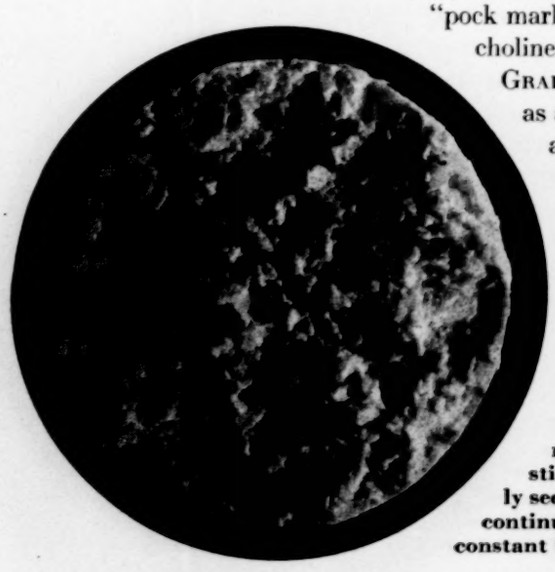

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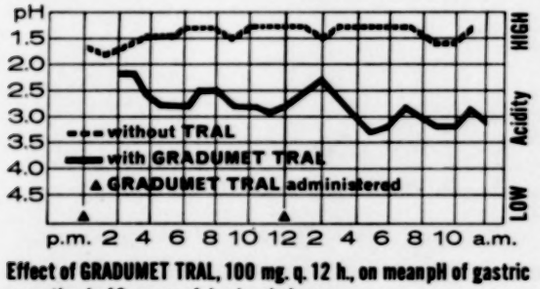
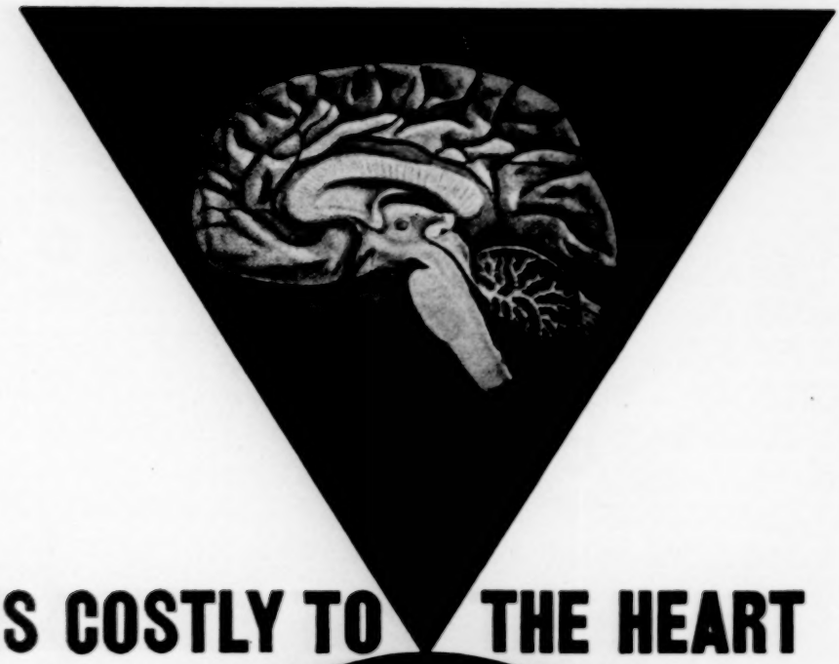


Effect of GRADUMET TRAL, 100 mg. q. 12 h., on mean pH of gastric secretion in 12 cases of duodenal ulcer.

[1]Kasich, A. M., and Fein, H. D.: "Effect of Hexocyclium Methosulfate, A New Anticholinergic Drug, In Conventional And Long-Acting Forms, Especially on the pH of Gastric Secretion As Studied in 48-Hour Gastric Analyses." Am. J. Digest. Dis., in press.

†TRAL TRADEMARK FOR HEXOCYCLIUM METHYLSULFATE, ABBOTT 801064

•GRADUMET TRADEMARK FOR LONG-RELEASE DOSAGE FORM, ABBOTT; PAT. APPLIED FOR.



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Emotional stress is a serious threat to an already diseased heart. Thoracic muscular tension simulating cardiac pain is an added source of apprehension.

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*Waldman, S. and Pelner, L.: *Management of anxiety associated with heart disease. Am. Pract. & Digest Treat.* 8:1075, July 1957.

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1. Carlozzi, M.: Antibiotic Med. & Clin. Therapy 5:146 (Feb.) 1958.

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Lerner, P. F.: Kemadrin, a New Drug for Treatment of Parkinsonian Disease, J. Nerv. & Ment. Dis. 123:79 (Jan.) 1956.

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*Masters, W.H.: Am. J. Obst. & Gynec., 74:733 (Oct.) 1957.

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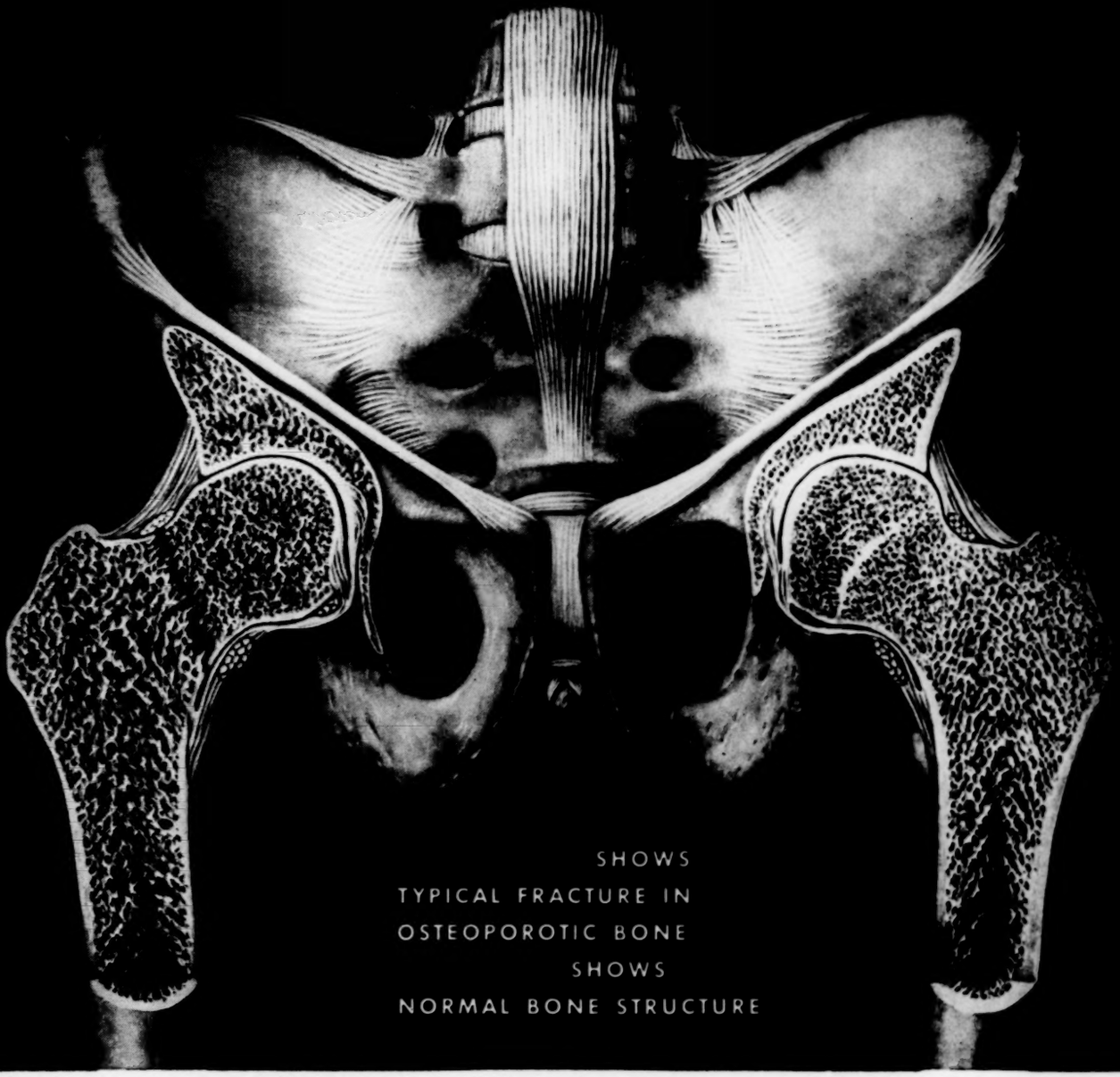

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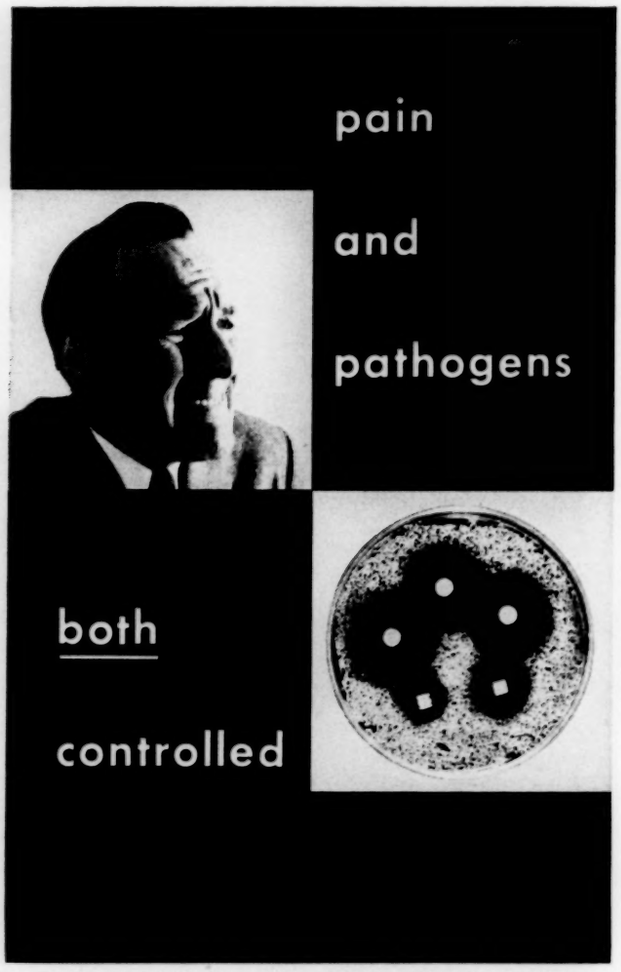

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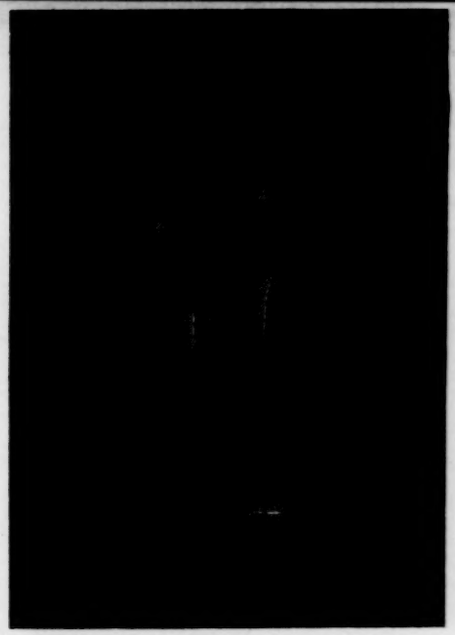

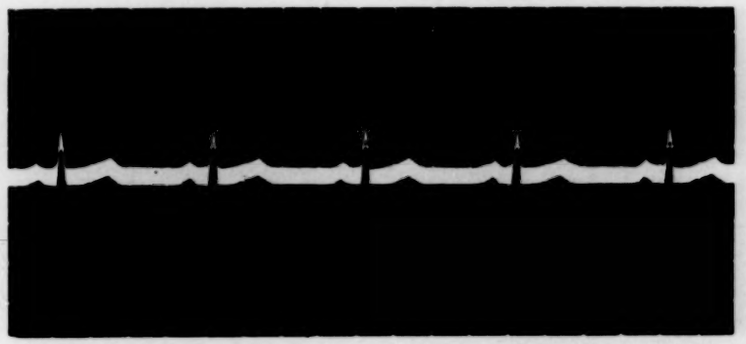

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1. Aravanis, C., and Luisada, A. A.: Am. J. Cardiology, in press.

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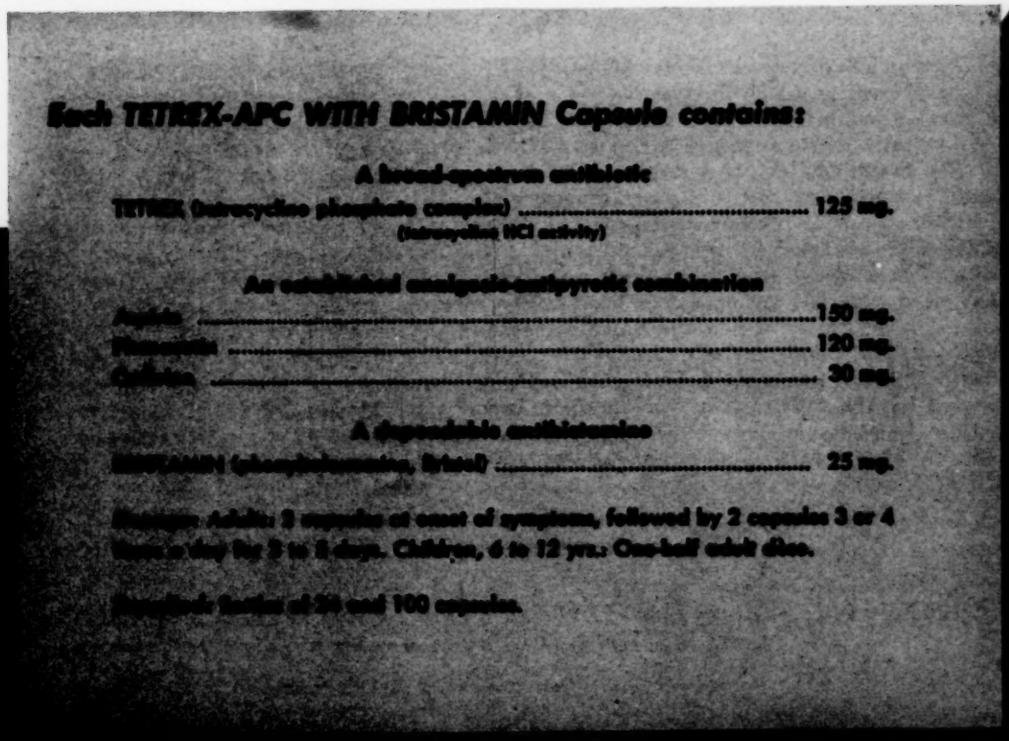

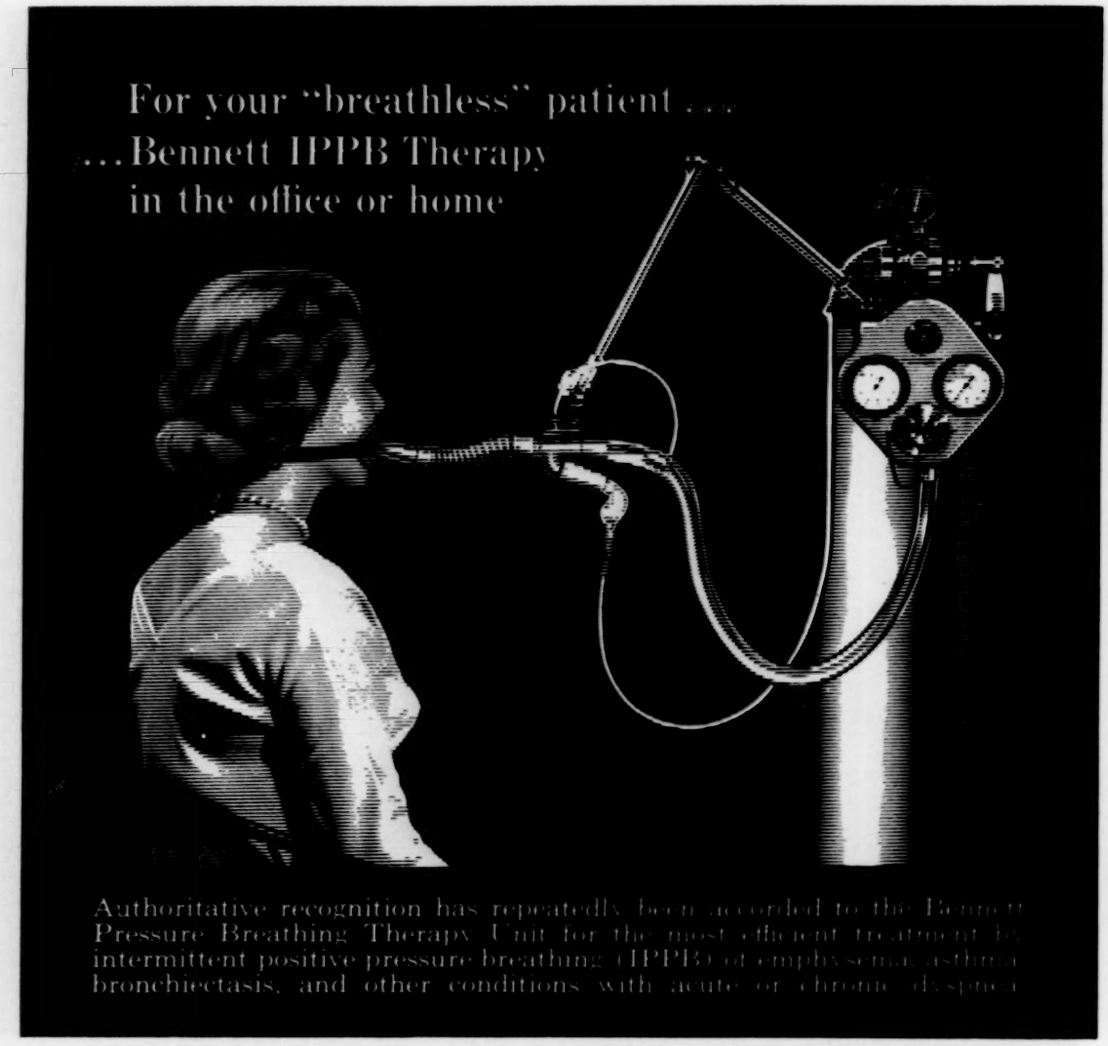

"Intermittent-positive-pressure breathing (IPPB), may be defined as a technique by which the lungs are actively inflated by means of a regulated, slightly positive pressure during inspiration, after which, during expiration, the lungs are passively deflated to atmospheric pressure, primarily by the elasticity of the lungs and the chest wall. The maximal peak pressure usually employed is that of 20 cm. of water, which is about one-fifth the maximal peak pressure that may be produced by a cough. The intermittent-positive-pressure breathing is produced by a type of respirator with a cycling valve operated by the gas pressure in the oxygen cylinder. A very slight inspiratory suction by the patient trips the cycling valve, and the positive pressure builds up during the inspiratory phase to the maximal peak pressure for which the regulator is set, after which the valve cycles and the pressure drops to atmospheric pressure during exhalation. The apparatus produces better depth of breathing, greater excursions of the diaphragm, and more uniform ventilation of the air sacs than that produced by the individual's own ambient breathing." Motley, H. L.: Evaluation of Function Impairment and New Developments in Therapy of Chronic Pulmonary Disease. A. M. A. Archives of Industrial Hygiene and Occupational Medicine, June, 1952, Vol. 5, pp 554-565.

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1. Corcoran, A. C., Lewis, L. A., Dustan, H. P., and Page, I. H.: Ann. New York Acad. Sc. *64*:620 (Nov. 16) 1956. 2. Perry, H. M., Jr., and Schroeder, H. A.: J. Chronic Dis. *2*:520 (Nov.) 1955. 3. Galioni, E. F.: California Med. *85*:97 (Aug.) 1956.

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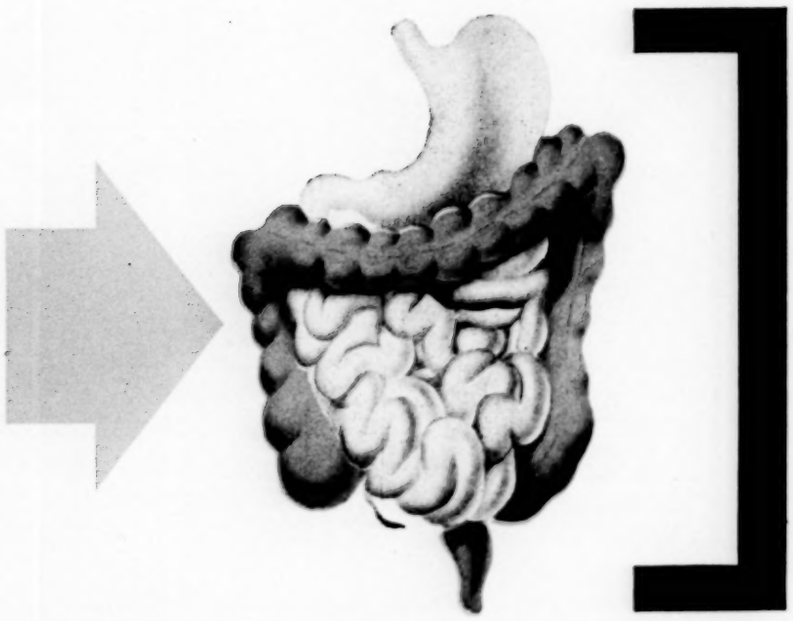

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(1) Kleckner, M. S., Jr.: J. Louisiana M. Soc. *108*:359, 1956. (2) Riese, J. A.: Am. J. Gastroenterol. *28*:541, 1957. (3) Settel, E.: J. Am. Geriatrics Soc. In press. (4) Jefferson, N. C., and Necheles, H.: J. Urol. *76*:651, 1956. (5) Necheles, H., and Kirshen, M. M.: The Physiologic Basis of Gastrointestinal Therapy, New York, Grune & Stratton, Inc., 1957, p. 88.

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1. Lewis, J.M., et al.: J. Pediat. 31:496.

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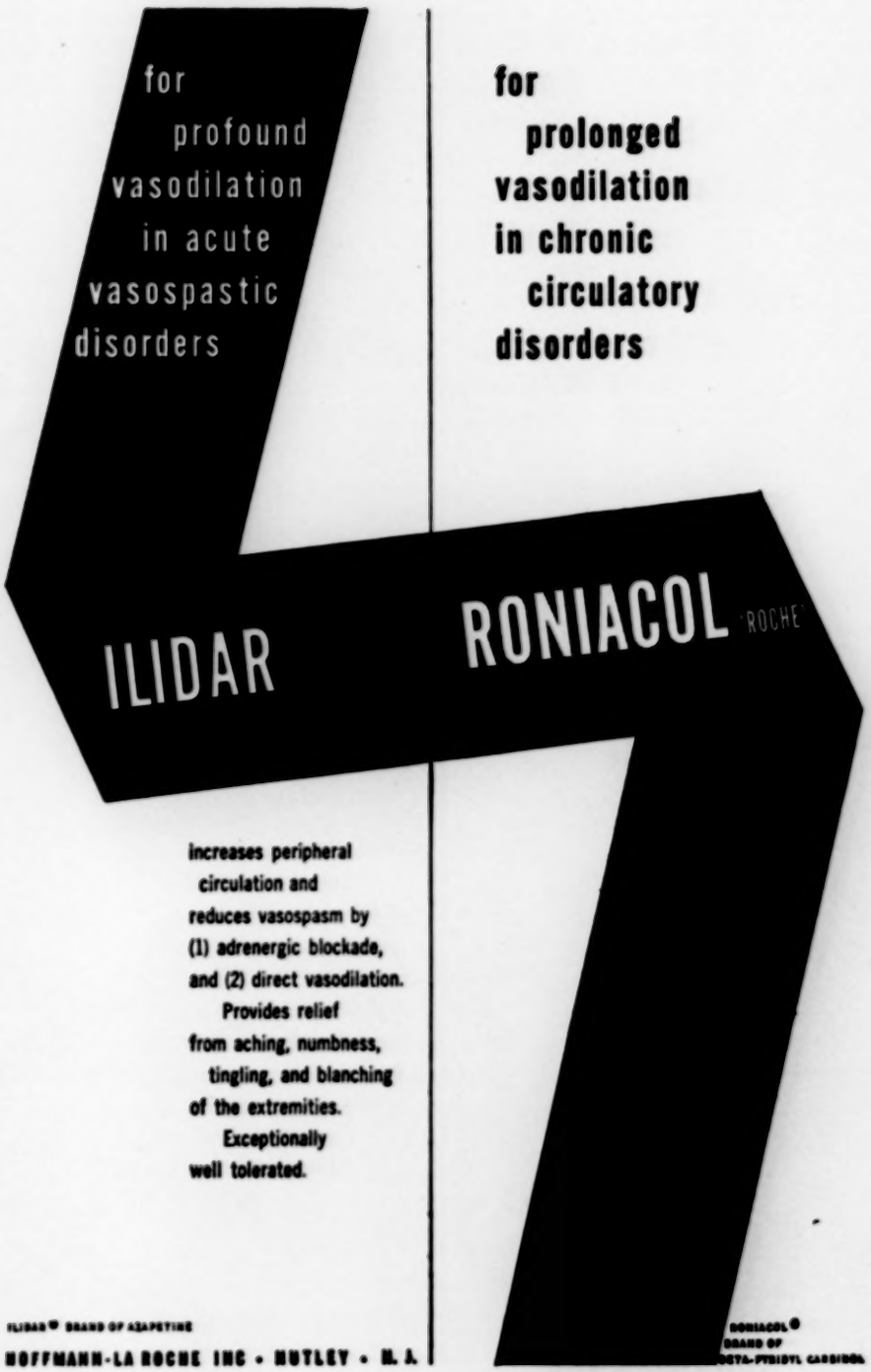
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1. Wright, W. T. Jr.; Pokorny, C., and Foster, T. L.: J. Kansas M. Soc. **57**:410, 1956.
2. Terman, L. A.: Illinois M.J. **3**:67, 1957.
3. Moyer, J. H.; Dennis, E., and Ford, R.: Arch. Int. Med. **96**:530, 1955.

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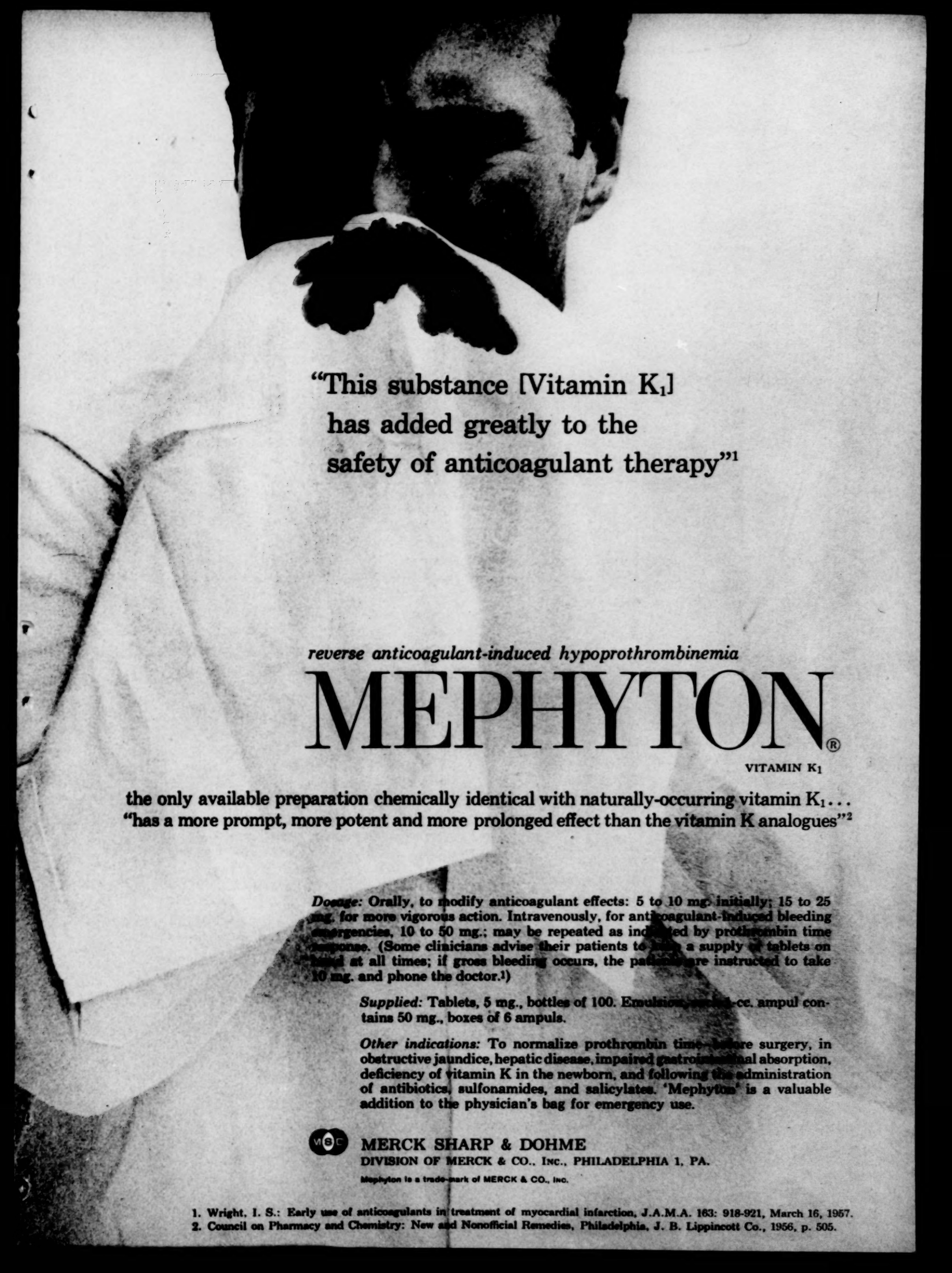

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1. Wright, I. S.: Early use of anticoagulants in treatment of myocardial infarction, J.A.M.A. 163: 918-921, March 16, 1957.
2. Council on Pharmacy and Chemistry: New and Nonofficial Remedies, Philadelphia, J. B. Lippincott Co., 1956, p. 505.

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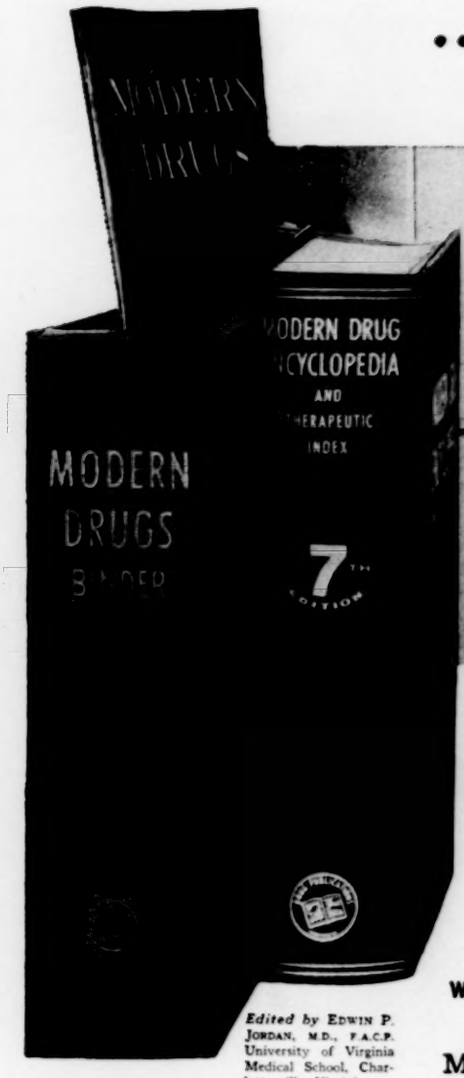

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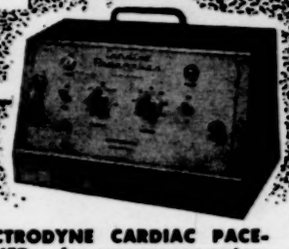
ELECTRODYNE PM-65* WITH ELECTRO-CARDIOSCOPE — detects and treats cardiac arrest automatically and externally — provides continuous visual display of electrocardiogram. (Scope optional).

ELECTRODYNE D-72 — for EXTERNALLY APPLIED treatment of ventricular fibrillation

These two important instruments are also available as one unit — Combination Pacemaker and Defibrillator. (Model 43)

ELECTRODYNE CARDIAC DEFIBRILLATOR — for emergency internal treatment of ventricular fibrillation. (Model 33)



ELECTRODYNE CARDIAC PACEMAKER — for emergency external treatment of ventricular standstill. (Model 27-A)

ELECTRODYNE E-11 — combination EXTERNAL Cardiac Pacemaker and EXTERNAL Defibrillator.

Cardiac arrest is certainly a serious occurrence in any hospital . . . that's why you should know the story about proven instruments for the detection and treatment of cardiac arrest and fibrillation.

From the introduction of the original and well known Cardiac Pacemaker, which was developed in conjunction with PAUL M. ZOLL, M.D., the Electrodyne Company has worked very closely with Dr. Zoll and his associates in continuous research and development in this specialized field of instrumentation.

Collectively these proven Electrodyne instruments represent an important family of life-saving medical equipment that is giving a feeling of security and peace of mind in the operating rooms and in the wards of hospitals throughout the world.

We will gladly send you complete literature upon request.

*** The need for continuous human observation is not required when the Electrodyne PM-65 is used in the detection and treatment of cardiac arrest.**

ELECTRODYNE CO., INC.

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NORWOOD, MASSACHUSETTS, U.S.A.

SPECIALISTS IN

ELECTRODYNE

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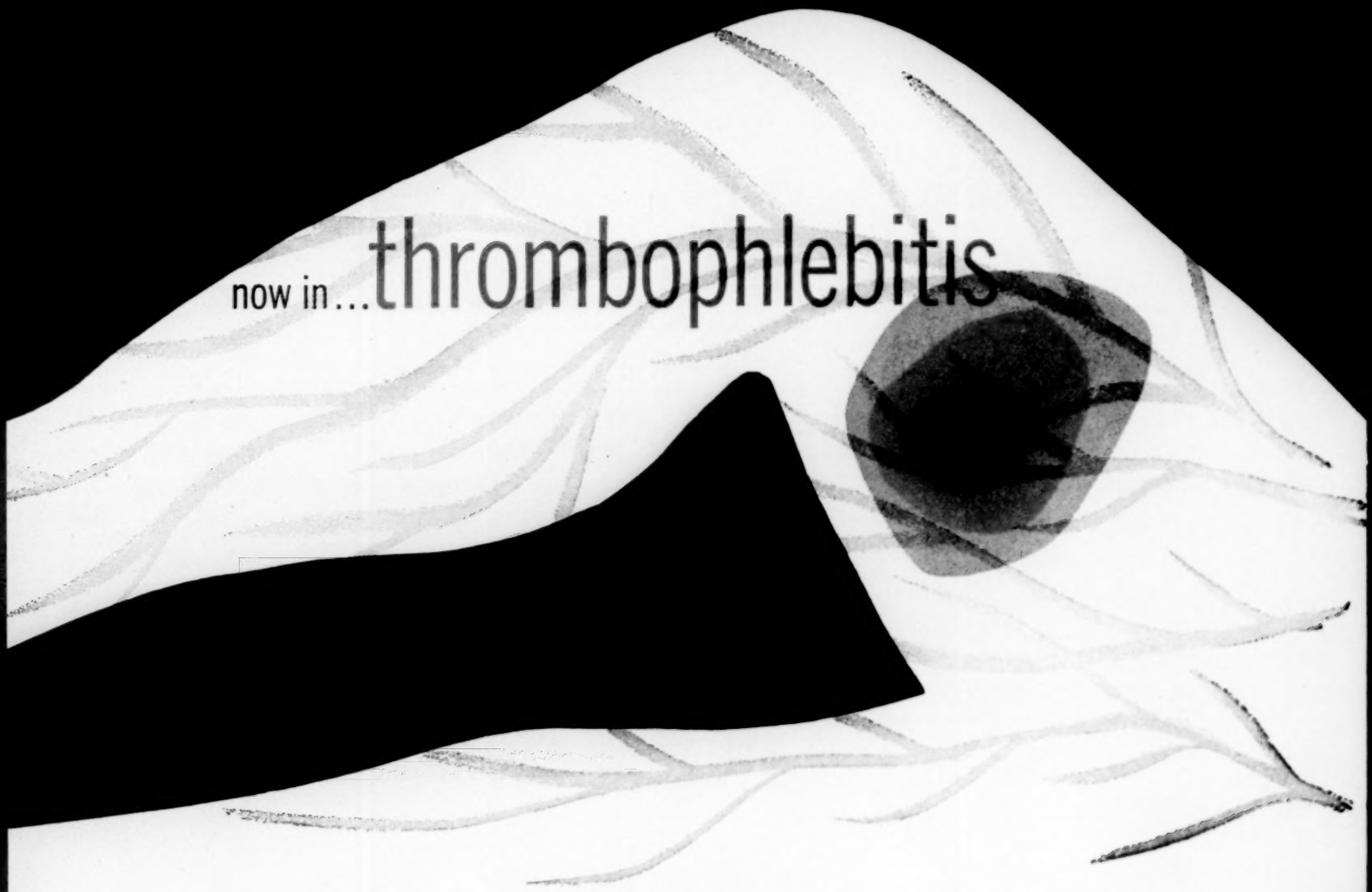

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"early and marked regression"[1]

in acute superficial thrombophlebitis

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(phenylbutazone GEIGY)

nonhormonal anti-inflammatory agent

Relieves Pain Rapidly—BUTAZOLIDIN usually produces complete relief of pain within 24 hours or less.[1,2]

Resolves Inflammation—Fever subsides and local heat, tenderness and swelling regress quickly.[1,3,4] "In the majority of cases there was complete resolution by the fourth day."[5]

Permits Early Ambulation—"As a rule within 24 hours, most patients were able to get up and walk about...."[1] This rapid response to BUTAZOLIDIN greatly reduces disability and economic loss for patients.

Short Course of Treatment—Most patients require only from 2 to 7 days' therapy.[1,5]

BUTAZOLIDIN® (phenylbutazone GEIGY). Red coated tablets of 100 mg. BUTAZOLIDIN Alka Capsules, each containing BUTAZOLIDIN 100 mg.; aluminum hydroxide 100 mg.; magnesium trisilicate 150 mg.; homatropine methylbromide 1.25 mg.

BUTAZOLIDIN being a potent therapeutic agent, physicians unfamiliar with it are urged to send for literature before instituting therapy.

GEIGY
Ardsley, New York

References: (1) Stein, I. D.: Circulation *12*:833, 1955. (2) Potvin, L.: Bull. Assoc. méd. lang. franç. Canada *85*:941, 1956. (3) Sigg, K.: Angiology *8*:44, 1957. (4) Elder, H. H. A., and Armstrong, J. B.: Practitioner *178*:479, 1957. (5) Braden, F. R.; Collins, C. G., and Sewell, J. W.: J. Louisiana M. Soc. *109*:372, 1957.

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No single drug can equal Tedral to protect

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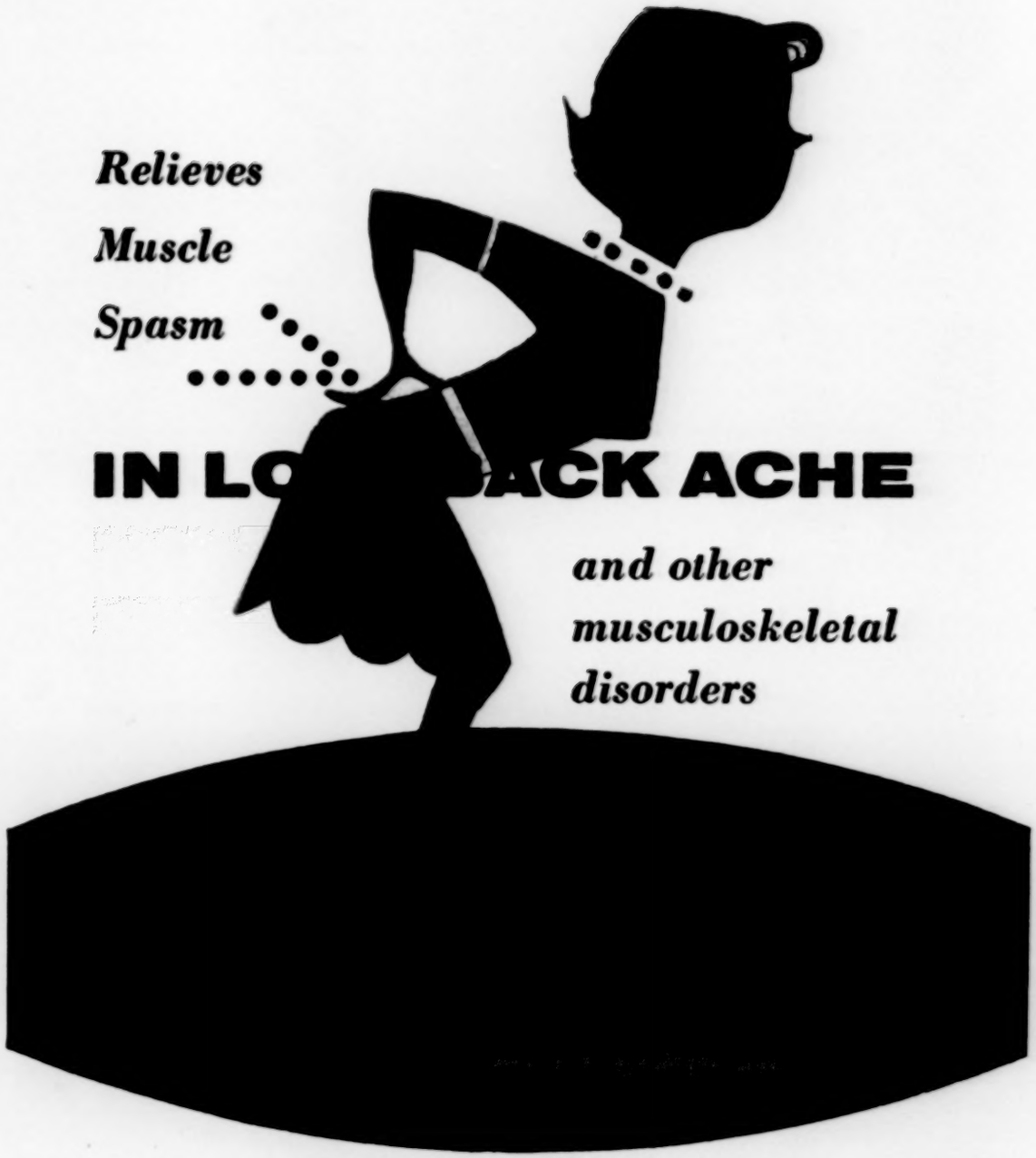
April, 1958



Relieves
Muscle
Spasm

IN LO[illegible]ACK ACHE

and other musculoskeletal disorders

In Parkinsonism

Highly selective action...energizing against weakness, fatigue, adynamia and akinesia...potent against sialorrhea, diaphoresis, oculogyria and blepharospasm... lessens rigidity and tremor... alleviates depression...safe... even in glaucoma.

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